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U.S. DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
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Carcinogenesis Testing Program
Division of Cancer Cause and Prevention
National Cancer Institute
National Institutes of Health
Bethesda, Maryland 20014

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REPORT ON THE BIOASSAY OF HYDRAZOBENZENE
FOR POSSIBLE CARCINOGENICITY

CARCINOGENESIS TESTING PROGRAM
DIVISION OF CANCER CAUSE AND PREVENTION
NATIONAL CANCER INSTITUTE, NATIONAL INSTITUTES OF HEALTH

FOREWORD: This report presents the results of the bioassay of hydrazobenzene conducted for the Carcinogenesis Testing Program, Division of Cancer Cause and Prevention, National Cancer Institute (NCI), National Institutes of Health, Bethesda, Maryland. This is one of a series of experiments designed to determine whether selected chemicals have the capacity to produce cancer in animals. Negative results, in which the test animals do not have a significantly greater incidence of cancer than control animals, do not necessarily mean the test chemical is not a carcinogen because the experiments are conducted under a limited set of circumstances. Positive results demonstrate that the test chemical is carcinogenic for animals under the conditions of the test and indicate a potential risk to man. The actual determination of the risk to man from animal carcinogens requires a wider analysis.

CONTRIBUTORS: This bioassay of hydrazobenzene was conducted by Mason Research Institute, Worcester, Massachusetts, initially under direct contract to the NCI and currently under a subcontract to Tracor Jitco, Inc., prime contractor for the NCI Carcinogenesis Testing Program.

The experimental design was determined by the NCI Project Officers, Dr. J. H. Weisburger (1,2) and Dr. E. K. Weisburger (1). The principal investigators for the contract were Dr. E. Smith (3) and Dr. A. Handler (3). Animal treatment and observation were supervised by Mr. G. Wade (3) and Ms. E. Zepp (3).

Histopathologic examinations were performed by Dr. Frederickson (3), Dr. D. W. Hayden (3), Dr. A. S. Krishna Murthy (3), and Dr. A. Russfield (3) at the Mason Research Institute, the pathology narratives were written by Dr. A. S. Krishna Murthy (3), and the diagnoses included in this report represent the interpretation of these pathologists. Histopathology findings and reports were reviewed by Dr. R. L. Schueler (4).

Compilation of individual animal survival, pathology, and summary tables was performed by EG&G Mason Research Institute (5); the statistical analysis was performed by Mr. W. W. Belew (6) using methods selected for the Carcinogenesis Testing Program by Dr. J. J. Gart (7).

This report was prepared at METREK, a Division of The MITRE Corporation (6) under the direction of the NCI. Those responsible for this report at METREK are the project coordinator, Dr. L. W. Thomas (6), task leader Dr. M. R. Kornreich (6), senior biologist Ms. P. Walker (6), biochemist Dr. B. Fuller (6), chemist Dr. N. Zimmerman (6), and technical editor Ms. P. A. Miller (6). The final report was reviewed by members of the participating organizations.

The following other scientists at the National Cancer Institute were responsible for evaluating the bioassay experiment, interpreting the results, and reporting the findings: Dr. K. C. Chu (1), Dr. C. Cueto, Jr. (1), Dr. J. F. Douglas (1), Dr. D. G. Goodman (1), Dr. R. A. Griesemer (1), Dr. H. A. Milman (1), Dr. T. W. Orme (1), Dr. R. A. Squire (1,8), Dr. J. M. Ward (1), and Dr. C. E. Whitmire (1).

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SUMMARY

A bioassay of technical-grade hydrazobenzene for possible carcinogenicity was conducted using Fischer 344 rats and B6C3F1 mice. Hydrazobenzene was administered in the feed, at either of two concentrations, to groups of 50 male and 47 to 50 female animals of each species. The time-weighted average dietary concentrations used in the rat bioassay were 0.008, 0.03, 0.004, and 0.01 percent for low dose males, high dose males, low dose females, and high dose females, respectively. The time-weighted average dietary concentrations used in the mouse bioassay were 0.008, 0.04, 0.004, and 0.04 percent for low dose males, high dose males, low dose females, and high dose females, respectively. After a 78-week period of compound administration, observation of the rats continued for an additional 28 to 30 weeks and observation of the mice continued for an additional 17 or 18 weeks. For each species, 47 to 50 animals of each sex were placed on test as controls.

In both species, adequate numbers of animals in all groups survived sufficiently long to be at risk from late-appearing tumors.

The incidence of hepatocellular carcinomas was significantly increased in dosed male rats and the incidence of neoplastic nodules of the liver was significantly increased in dosed female rats. A significant increase in the combined incidence of squamous-cell carcinomas or squamous-cell papillomas of the Zymbal's gland, the ear canal, or the skin of the ear was observed among high dose male rats. A significant increase in mammary adenocarcinomas was observed among dosed female rats.

The incidence of hepatocellular carcinomas was significantly increased among female mice, but no significant increase in liver tumors was observed among male mice.

Under the conditions of this bioassay, hydrazobenzene was carcinogenic to Fischer 344 rats of both sexes, causing increased incidences of hepatocellular carcinoma and Zymbal's gland squamous-cell neoplasms in male rats, neoplastic nodules of the liver in female rats, and mammary adenocarcinomas in female rats. Hydrazobenzene was also carcinogenic to female B6C3F1 mice, causing an increased incidence of hepatocellular carcinomas. The compound was not carcinogenic to male B6C3F1 mice.

TABLE OF CONTENTS

LIST OF ILLUSTRATIONS

LIST OF TABLES

LIST OF TABLES (Concluded)

I. INTRODUCTION

Hydrazobenzene (NCI No. C01854) is a hydrazine derivative selected for bioassay by the National Cancer Institute because of the documented carcinogenicity of the parent compound hydrazine and of certain substituted hydrazines (International Agency for Research on Cancer, 1974). The increased incidence of bladder cancer observed among workers in the dye manufacturing industry (Wynder et al., 1963; Anthony and Thomas, 1970) was also a factor in the selection of this chemical for testing.

The Chemical Abstracts Service (CAS) Ninth Collective Index* (1977) name for this compound is 1,2-diphenylhydrazine. It is also known as symmetrical (sym-) diphenylhydrazine (Weast, 1972).

Treatment of hydrazobenzene with hot mineral acid results in the production of benzidine (the so-called "benzidine rearrangement") and hydrazobenzene finds application in the dye manufacturing industry as a precursor of this important dye intermediate and potent carcinogen. An additional use of hydrazobenzene is in the preparation of phenylbutazone (via condensation with diethylbutyl malonate), a widely used agent against rheumatoid arthritis and related conditions (Wenner, 1967).

Although specific production figures are not available, the listing of hydrazobenzene in the 1977 Directory of Chemical Producers, U.S.A. (Stanford Research Institute, 1977) implies an annual commercial

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*The CAS registry number is 122-66-7.

production in excess of 1000 pounds or $1000 in value. This production would appear to be in addition to the quantities produced at dye manufacturing facilities by, for example, the reduction of nitrobenzene with zinc dust and sodium hydroxide prior to the benzidine rearrangement (Bannister, 1967).

The potential for exposure to hydrazobenzene is greatest for workers in the dye manufacturing industry although workers in the pharmaceutical industry may also experience contact with the chemical

Hydrazine derivatives tend to be local irritants, convulsants, hepatotoxins and hemolytic agents and are absorbed by all routes of administration (Sutton, 1967). It has been suggested that benzidine may be produced from hydrazobenzene, following ingestion, by acidity in the stomach (International Agency for Research on Cancer, 1972).

II. MATERIALS AND METHODS

A. Chemicals

Technical-grade hydrazobenzene (Figure 1) was purchased from K & K Labs. Chemical analysis was performed by Mason Research Institute, Worcester, Massachusetts. The experimentally determined melting point of 120° to 124°C suggested the presence of impurities due to its difference from the literature value of 129° to 131°C. Thin-layer chromatography showed a major spot with an Rf of 0.63 and a single unidentified minor spot indicating the presence of at least one impurity. Infrared analysis was not inconsistent with the structure of hydrazobenzene.

Throughout this report the term hydrazobenzene is used to represent this technical-grade material.

B. Dietary Preparation

The basal laboratory diet for both treated and control animals was Wayne Lab-Blox® (Allied Mills, Inc., Chicago, Illinois). Hydrazobenzene was administered to the treated animals as a component of the diet. The hydrazobenzene was first ground to a powder in a Quaker City Crystal Mill and then returned to the original metal container. To prepare the chemical diet mixture, the chemical was removed from the stock container, weighed out in proper amounts for dosage preparation, and hand-blended in an aluminum bowl with an aliquot of ground feed. Once visual homogeneity was attained, the mixture was placed into a 6 kg capacity Patterson-Kelley standard

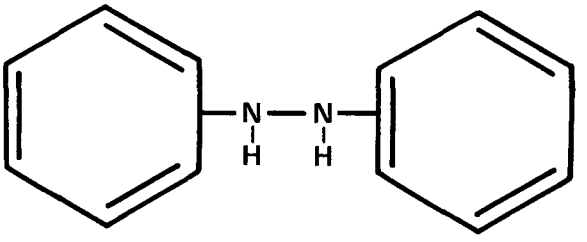


FIGURE 1
CHEMICAL STRUCTURE OF HYDRAZOBENZENE

model twin-shell stainless steel V-blender with the remainder of the diet. After 20 minutes of blending, the mixtures were placed in double plastic bags and stored in the dark at 4°C. Mixtures were prepared once weekly and stored for a maximum period of one week.

C. Animals

Two animal species, rats and mice, were used in the carcinogenicity bioassay. Fischer 344 rats and B6C3F1 mice were obtained through contracts of the Division of Cancer Treatment, National Cancer Institute. High dose rats and their controls and all mice were supplied by Charles River Breeding Laboratories, Inc., Wilmington, Massachusetts. Low dose rats and their controls were supplied by Laboratory Supply Company, Inc., Indianapolis, Indiana. Animals to be treated were received separately from their respective control groups, except low dose treated and control rats, which were received on the same date. Upon arrival, a sample of animals was examined for parasites and other signs of disease. The remaining animals were quarantined by species for 2 weeks prior to initiation of test. Animals were assigned to groups and distributed among cages so that the average body weight per cage was approximately equal for a given species and sex.

D. Animal Maintenance

All animals were housed by species in rooms having a temperature range of 23° to 34°C. Incoming air was filtered through Tri-Dek® 15/40 denier Dacron® filters (Tri-Dim Filter Corp., Hawthorne, New

Jersey) providing six changes of room air per hour. Fluorescent lighting was provided on a 12-hour-daily cycle.

Rats were housed five per cage by sex. During quarantine and for the first 13 months of study, all rats were kept in galvanized- or stainless-steel wire-mesh cages suspended above newspapers. Newspapers under cages were replaced daily and cages and racks washed weekly. For the remainder of the study, high dose, low dose, and control rats were housed in suspended polycarbonate cages equipped with nonwoven fiber filter sheets. Clean bedding and cages were provided twice weekly. Corncob bedding (SAN-I-CEL®, Paxton Processing Company, Paxton, Illinois) was used for low dose treated and control rats for the first 9 months that they were housed in polycarbonate cages, and Aspen hardwood chip bedding (American Excelsior Company, Baltimore, Maryland) was used for the remainder of the study. High dose treated and control rats received Aspen bedding for the entire time that they were housed in polycarbonate cages. Stainless steel cage racks were cleaned once every two weeks, and disposable filters were replaced at that time.

Mice were housed by sex in polycarbonate cages. During quarantine and periods of compound administration, cages were fitted with perforated stainless steel lids. During the untreated observation period, stainless steel wire bar lids were used. Both types of lids were from Lab Products, Inc., Garfield, New Jersey. Nonwoven fiber filter bonnets were used over cage lids. Low dose mice and their

controls were housed ten per cage for the first 16 months of study and five per cage thereafter. High dose mice and their controls were housed ten per cage initially and then five per cage after 11 months. Cages, lids, filters, and bedding were provided three times per week when cage populations were ten and twice per week when cage populations were reduced to five. Hardwood chip bedding (Ab-sorb-dri®, Wilner Wood Products Company, Norway, Maine) was used for the first 30 weeks for low dose and low dose control mice, for the first 6 weeks for high dose mice, and for 2 weeks for high dose control mice. SAN-I-CEL® corncob bedding was used for all mice for the next 12 months. For the remainder of the study, Aspen bedding was used. Reusable filter bonnets and pipe cage racks were sanitized every 2 weeks throughout the study.

Water was available for both species from 250 ml water bottles equipped with rubber stoppers and stainless steel sipper tubes. Bottles were replaced twice weekly and, for rats only, water was supplied as needed between changes. Food and water were available ad libitum.

Pelleted Wayne Lab-Blox® was fed to low dose rats and their controls during quarantine and to all rats and mice during the untreated observation period. During the dosing period, all treated animals were fed Wayne Lab-Blox® meal containing the appropriate concentration of hydrazobenzene. Control animals had untreated meal available. Alpine® aluminum feed cups (Curtin Matheson Scientific,

Inc., Woburn, Massachusetts) containing stainless steel baffles were used to distribute powdered feed to all mice and to low dose rats and their controls throughout the study. High dose treated and control rats were fed from Alpine® feed cups during quarantine and for the first 11 months of study. For the remainder of the study, high dose treated and control rats were fed from stainless steel gangstyle feed hoppers (Scientific Cages, Inc., Bryan, Texas). During the final observation period, mice were fed pellets from a wire bar hopper incorporated into the cage lid, and rats were fed pellets on the cage floor. Food hoppers were changed on the same schedule as were cages. Food was replenished daily in Alpine® feed cups.

Low dose rats and their controls and high dose rats were in a room with other rats receiving diets containing* 5-nitro-o-toluidine (99-55-8); 2-aminoanthraquinone (117-79-3); 3-amino-9-ethylcarbazole hydrochloride; 6-nitrobenzimidazole (94-52-0); 1-nitronaphthalene (86-57-7); 2,4-diaminoanisole sulfate (615-05-4); and APC (8003-03-0). High dose control rats were in a room with other rats receiving diets containing amitrole (61-82-5); 2-methyl-1-nitroanthraquinone (129-15-7); and 3-nitro-p-acetophenetide (1777-84-0).

Low dose mice were housed in a room with other mice receiving diets containing 2,5-toluenediamine sulfate (6369-59-1); 5-nitro-o-toluidine (99-55-8); 3-amino-9-ethylcarbazole hydrochloride; 6-nitrobenzimidazole (94-52-0); 5-nitro-o-anisidine (99-59-2); 1-nitronaphthalene (86-57-7);

*CAS registry numbers are given in parentheses.

and 2,4-diaminoanisole sulfate (615-05-4). High dose mice were housed in a room in which other mice were receiving diets containing 2,3,5,6-tetrachloro-4-nitroanisole (2438-88-2); tris(2,3-dibromopropyl) phosphate (126-72-7); N-(1-naphthyl)ethylenediamine dihydrochloride (1465-25-4); 2-chloro-p-phenylenediamine sulfate (61702-44-1); and acetone (67-64-1). All control mice were in a room where other mice were receiving diets containing amitrole (61-82-5); N,N-dimethyl-p-nitrosoaniline (138-89-6); 2,5-toluenediamine sulfate (6369-59-1); 2,4-dinitrotoluene (121-14-2); 2-aminoanthraquinone (117-79-3); 3-amino-4-ethoxyacetanilide (17026-81-2); 3-amino-9-ethylcarbazole hydrochloride; 1-amino-2-methylanthraquinone (82-28-0); 5-nitro-o-anisidine (99-59-2); 4-nitroanthranilic acid (619-17-0); 1-nitro-naphthalene (86-57-7); 2,4-diaminoanisole sulfate (615-05-4); and APC (8003-03-0).

E. Selection of Initial Concentrations

In order to establish the maximum tolerated concentrations of hydrazobenzene for administration to treated animals in the chronic studies, subchronic toxicity tests were conducted with both rats and mice. Animals of each species were distributed among several groups, each consisting of five males and five females. The chemical was incorporated into the laboratory diet and supplied ad libitum to the rat and mouse groups for a total of 4 weeks. This dosing period was followed by a 2-week observation period during which animals were

fed the basal diet. Eight of nine male rat groups received concentrations of 0.007, 0.014, 0.028, 0.055, 0.108, 0.214, 0.301 and 0.423 percent. The ninth male rat group served as a control, receiving only the basal laboratory diet. Nine of ten female rat groups were fed dietary concentrations of 0.00008, 0.0003, 0.0011, 0.002, 0.004, 0.015, 0.104, 0.731, and 5.138 percent. The tenth group served as a control group.

Eight of the nine male mouse groups were given dietary concentrations of 0.007, 0.014, 0.028, 0.055, 0.108, 0.214, 0.301, and 0.423 percent. The ninth group served as a control, receiving only the basal laboratory diet. Nine of the ten female mouse groups were given dietary concentrations of 0.0003, 0.0008, 0.0011, 0.002, 0.004, 0.015, 0.104, 0.731, and 5.138 percent. The tenth group served as a control.

The highest concentration causing no deaths, no compound-related gross abnormalities, and no mean group body weight depression in excess of 11 percent relative to controls during the 6-week subchronic test was selected as the high concentration utilized for the rat and mouse chronic bioassays.

Two of the five male rats receiving concentrations of 0.108 percent died. All rats receiving higher concentrations died. One male mouse receiving a concentration of 0.301 percent, two male mice receiving 0.423 percent, four female mice receiving 0.731 percent, and all female mice receiving 5.138 percent died. Mean body weight depression

patterns were not consistent. Intestinal hemorrhage was the single gross abnormality consistently observed in these mice.

The initial high dietary concentrations used in the chronic study were 0.03 percent for male rats, 0.01 percent for female rats, and 0.04 percent for male and female mice.

F. Experimental Design

The experimental design parameters for the chronic study (species, sex, group size, concentrations administered, duration of treated and untreated observation periods, and time-weighted average concentrations) are summarized in Tables 1 and 2.

Male rats were all approximately 6 weeks old at the time they were placed on test. The initial dietary concentrations of hydrazobenzene were 0.007 and 0.0035 percent. After week 9, the higher of these doses was raised to 0.008 percent. For male rats the group receiving the lower dose (0.0035 percent) was sacrificed after 41 weeks because the dose level was considered, on the basis of weight depression, to be inadequate. No histopathologic examinations of these animals were performed. A new group, receiving 0.03 percent, was started with its own control group. Throughout this report, those male rats receiving 0.03 percent hydrazobenzene and their controls are referred to as the high dose and high dose control groups, respectively. Throughout this report, those male rats initially receiving a concentration of 0.007 percent and their controls are referred to as the low dose and low dose control groups, respectively.

TABLE 1

DESIGN SUMMARY FOR FISCHER 344 RATS
HYDRAZOBENZENE FEEDING EXPERIMENT

	INITIAL GROUP SIZE	HYDRAZOBENZENE CONCENTRATION (PERCENT)	OBSERVATION PERIOD TREATED (WEEKS)	OBSERVATION PERIOD UNTREATED (WEEKS)	TIME-WEIGHTED AVERAGE CONCENTRATION[a]
MALE					
LOW DOSE CONTROL	50	0	0	108	0
HIGH DOSE CONTROL	49	0	0	109	0
LOW DOSE	50	0.007 0.008 0	9 69	 29	0.008
HIGH DOSE	50	0.03 0	78	 28	0.03
FEMALE					
LOW DOSE CONTROL	50	0	0	109	0
HIGH DOSE CONTROL	50	0	0	109	0
LOW DOSE	50	0.004 0	78	 30	0.004
HIGH DOSE	50	0.01 0	78	 29	0.01

[a] Time-weighted average concentration = $\frac{\Sigma(\text{concentration X weeks received})}{\Sigma(\text{weeks receiving chemical})}$

TABLE 2

DESIGN SUMMARY FOR B6C3F1 MICE
HYDRAZOBENZENE FEEDING EXPERIMENT

	INITIAL GROUP SIZE	HYDRAZOBENZENE CONCENTRATION (PERCENT)	OBSERVATION PERIOD TREATED (WEEKS)	OBSERVATION PERIOD UNTREATED (WEEKS)	TIME-WEIGHTED AVERAGE CONCENTRATION[a]
MALE					
LOW DOSE CONTROL	50	0	0	95	0
HIGH DOSE CONTROL	50	0	0	96	0
LOW DOSE	50	0.007 0.008 0	9 69	 17	0.008
HIGH DOSE	50	0.04 0	78	 17	0.04
FEMALE					
LOW DOSE CONTROL	50	0	0	96	0
HIGH DOSE CONTROL	50	0	0	96	0
LOW DOSE	47	0.004 0	78	 17	0.004
HIGH DOSE	50	0.04 0	78	 18	0.04

[a] Time-weighted average concentration = $\frac{\Sigma(\text{concentration X weeks received})}{\Sigma(\text{weeks receiving chemical})}$

Female rats were all approximately 6 weeks old at the time they were placed on test. The initial dietary concentrations of hydrazobenzene were 0.004 and 0.002 percent. The female rats receiving 0.002 percent were sacrificed after 41 weeks because the dose level was considered, on the basis of weight depression, to be inadequate. No histopathologic examinations of these animals were performed. A new dosed group, receiving 0.01 percent, was started with a new control. Throughout this report, those female rats receiving 0.01 percent and their controls are referred to as the high dose and high dose control groups, respectively. Throughout this report those female rats initially receiving a concentration of 0.004 percent and their controls are referred to as the low dose and low dose control groups, respectively.

Male mice were all approximately 6 weeks old at the time they were placed on test. The initial dietary concentrations of hydrazobenzene were 0.007 and 0.0035 percent. After week 9, the higher dose was increased to 0.008 percent. The male mice initially receiving 0.0035 percent were sacrificed after 22 weeks because the dose level was considered, on the basis of weight depression, to be inadequate. No histopathologic examinations of these animals were performed. A new dosed group, receiving 0.04 percent, was started with a new control group. Throughout this report, those male mice receiving 0.04 percent and their controls are referred to as the high dose and high dose control groups, respectively. Throughout this report those

male mice initially receiving a concentration of 0.007 percent and their controls are referred to as the low dose and low dose control groups, respectively.

Female mice were all approximately 6 weeks old at the time they were placed on test. The initial dietary concentrations of hydrazobenzene were 0.004 and 0.002 percent. The female mice initially receiving 0.002 percent were sacrificed after 22 weeks because the dose level was considered, on the basis of weight depression, to be inadequate. No histopathologic examinations of these animals were performed. A new dosed group, receiving 0.04 percent, was started with a new control group. Throughout this report, those female mice receiving a concentration of 0.04 percent and their controls are referred to as the high dose and high dose control groups, respectively. Throughout this report those female mice initially receiving a concentration of 0.004 percent and their controls are referred to as the low dose and low dose control groups, respectively.

Treated rats and mice (except for groups terminated early) were supplied with dosed feed for a total of 78 weeks followed by a 26-week observation period for rats and a 13-week observation period for mice.

G. Clinical and Histopathologic Examinations

Animals were weighed immediately prior to initiation of the experiment. Body weights were recorded twice weekly for the first 12 weeks of the study and at monthly intervals thereafter. From the first day, all animals were inspected twice daily for mortality.

Food consumption, for two cages from each group, was monitored for seven consecutive days once a month for the first nine months of the bioassay and for three consecutive days each month thereafter. The presence of tissue masses and lesions was determined by monthly observation and palpation of each animal.

A necropsy was performed on each animal regardless of whether it died, was killed when moribund, or was sacrificed at the end of the bioassay. The animals were euthanized by carbon dioxide inhalation, and were immediately necropsied. The histopathologic examination consisted of gross and microscopic examination of major tissues, organs, and gross lesions taken from sacrificed animals and, whenever possible, from animals found dead.

Tissues were preserved in 10 percent buffered formalin, embedded in paraffin, sectioned, and stained with hematoxylin and eosin prior to microscopic examination. An occasional section was subjected to special staining techniques for more definitive diagnosis.

Slides were prepared from the following tissues: skin, subcutaneous tissue, lungs and bronchi, trachea, bone marrow, spleen, lymph nodes, thymus, heart, salivary gland, liver, gallbladder (mice), pancreas, esophagus, stomach, small intestine, large intestine, kidney, urinary bladder, seminal vesicle, pituitary, adrenal, thyroid, parathyroid, ear, testis, prostate, brain, Zymbal's gland, uterus, mammary gland, and ovary.

A few tissues were not examined for some animals, particularly for those that died early. Also, some animals were missing, cannibalized, or judged to be in such an advanced state of autolysis as to preclude histopathologic interpretation. Thus, the number of animals for which particular organs, tissues, or lesions were examined microscopically varies and does not necessarily represent the number of animals that were placed on experiment in each group.

H. Data Recording and Statistical Analyses

Pertinent data on this experiment have been recorded in an automatic data processing system, the Carcinogenesis Bioassay Data System (Linhart et al., 1974). The data elements include descriptive information on the chemicals, animals, experimental design, clinical observations, survival, body weight, and individual pathologic results, as recommended by the International Union Against Cancer (Berenblum, 1969). Data tables were generated for verification of data transcription and for statistical review.

These data were analyzed using the statistical techniques described in this section. Those analyses of the experimental results that bear on the possibility of carcinogenicity are discussed in the statistical narrative sections.

Probabilities of survival were estimated by the product-limit procedure of Kaplan and Meier (1958) and are presented in this report in the form of graphs. Animals were statistically censored as of the time that they died of other than natural causes or were found to be

missing; animals dying from natural causes were not statistically censored. Statistical analyses for a possible dose-related effect on survival used the method of Cox (1972) when testing two groups for equality and used Tarone's (1975) extensions of Cox's methods when testing a dose-related trend. One-tailed P-values have been reported for all tests except the departure from linearity test, which is only reported when its two-tailed P-value is less than 0.05.

The incidence of neoplastic or nonneoplastic lesions has been given as the ratio of the number of animals bearing such lesions at a specific anatomic site (numerator) to the number of animals in which that site was examined (denominator). In most instances, the denominators included only those animals for which that site was examined histologically. However, when macroscopic examination was required to detect lesions prior to histologic sampling (e.g., skin or mammary tumors), or when lesions could have appeared at multiple sites (e.g., lymphomas), the denominators consist of the numbers of animals necropsied.

The purpose of the statistical analyses of tumor incidence is to determine whether animals receiving the test chemical developed a significantly higher proportion of tumors than did the control animals. As a part of these analyses, the one-tailed Fisher exact test (Cox, 1970, pp. 48-52) was used to compare the tumor incidence of a control group to that of a group of treated animals at each dose level. When results for a number of treated groups, k, are compared simultaneously

with those for a control group, a correction to ensure an overall significance level of 0.05 may be made. The Bonferroni inequality (Miller, 1966, pp. 6-10) requires that the P-value for any comparison be less than or equal to 0.05/k. In cases where this correction was used, it is discussed in the narrative section. It is not, however, presented in the tables, where the Fisher exact P-values are shown.

The Cochran-Armitage test for linear trend in proportions, with continuity correction (Armitage, 1971, pp. 362-365), was also used when appropriate. Under the assumption of a linear trend, this test determined if the slope of the dose-response curve is different from zero at the one-tailed 0.05 level of significance. Unless otherwise noted, the direction of the significant trend was a positive dose relationship. This method also provides a two-tailed test of departure from linear trend.

A time-adjusted analysis was applied when numerous early deaths resulted from causes that were not associated with the formation of tumors. In this analysis, deaths that occurred before the first tumor was observed were excluded by basing the statistical tests on animals that survived at least 52 weeks, unless a tumor was found at the anatomic site of interest before week 52. When such an early tumor was found, comparisons were based exclusively on animals that survived at least as long as the animal in which the first tumor was found. Once this reduced set of data was obtained, the standard

procedures for analyses of the incidence of tumors (Fisher exact tests, Cochran-Armitage tests, etc.) were followed.

When appropriate, life-table methods were used to analyze the incidence of tumors. Curves of the proportions surviving without an observed tumor were computed as in Saffiotti et al. (1972). The week during which animals died naturally or were sacrificed was entered as the time point of tumor observation. Cox's methods of comparing these curves were used for two groups; Tarone's extension to testing for linear trend was used for three groups. The statistical tests for the incidence of tumors which used life-table methods were one-tailed and, unless otherwise noted, in the direction of a positive dose relationship. Significant departures from linearity ($P < 0.05$, two-tailed test) were also noted.

The approximate 95 percent confidence interval for the relative risk of each dosed group compared to its control was calculated from the exact interval on the odds ratio (Gart, 1971). The relative risk is defined as p_t/p_c where p_t is the true binomial probability of the incidence of a specific type of tumor in a treated group of animals and p_c is the true probability of the spontaneous incidence of the same type of tumor in a control group. The hypothesis of equality between the true proportion of a specific tumor in a treated group and the proportion in a control group corresponds to a relative risk of unity. Values in excess of unity represent the condition of a larger proportion in the treated group than in the control.

The lower and upper limits of the confidence interval of the relative risk have been included in the tables of statistical analyses. The interpretation of the limits is that in approximately 95 percent of a large number of identical experiments, the true ratio of the risk in a treated group of animals to that in a control group would be within the interval calculated from the experiment. When the lower limit of the confidence interval is greater than one, it can be inferred that a statistically significant result (a $P < 0.025$ one-tailed test when the control incidence is not zero, $P < 0.050$ when the control incidence is zero) has occurred. When the lower limit is less than unity but the upper limit is greater than unity, the lower limit indicates the absence of a significant result while the upper limit indicates that there is a theoretical possibility of the induction of tumors by the test chemical which could not be detected under the conditions of this test.

III. CHRONIC TESTING RESULTS: RATS

A. Body Weights and Clinical Observations

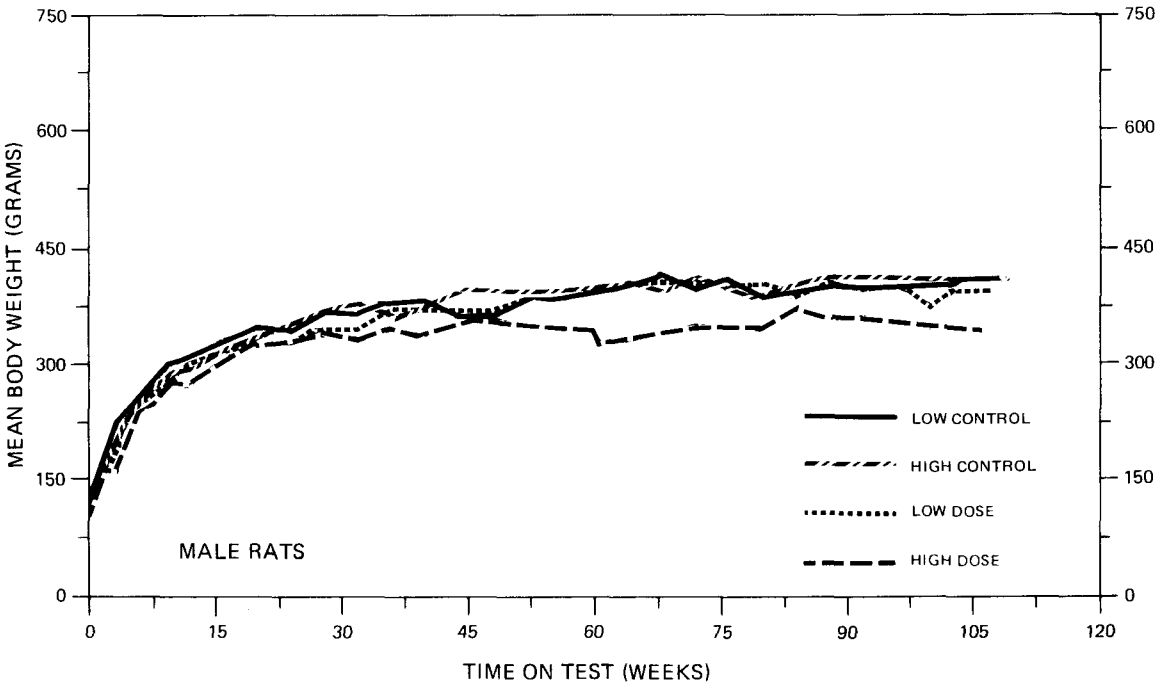
During this bioassay, slight depression of mean group body weight relative to controls was apparent for high dose male rats but not for low dose male rats. Among female rats, a slight depression of mean group body weight was observed for the low dose group after week 46 and for the high dose group after week 22 (Figure 2).

Palpable subcutaneous masses were the only frequent clinical observation. They were observed in ten high dose control females, two high dose control males, three high dose females and two low dose males. One high dose male had a firm nodule on the ear. Sporadic, isolated observations included: One high dose male with an eye infection, one high dose control female with eye discoloration and another with exopthalmia, one low dose control male with an encrusted lesion on the dorsal surface, a brown vaginal exudate in one high dose female, and alopecia in one high dose control female. No other clinical abnormalities were recorded for male or female rats.

B. Survival

The estimated probabilities of survival for male and female rats in the control and hydrazobenzene-dosed groups are shown in Figure 3.

For male rats the Cox tests did not indicate positive associations between increased dosage and accelerated mortality. From each of the treated groups and from the untreated controls, five rats were sacrificed in week 78. In week 100, 64 percent (32/50) of the high



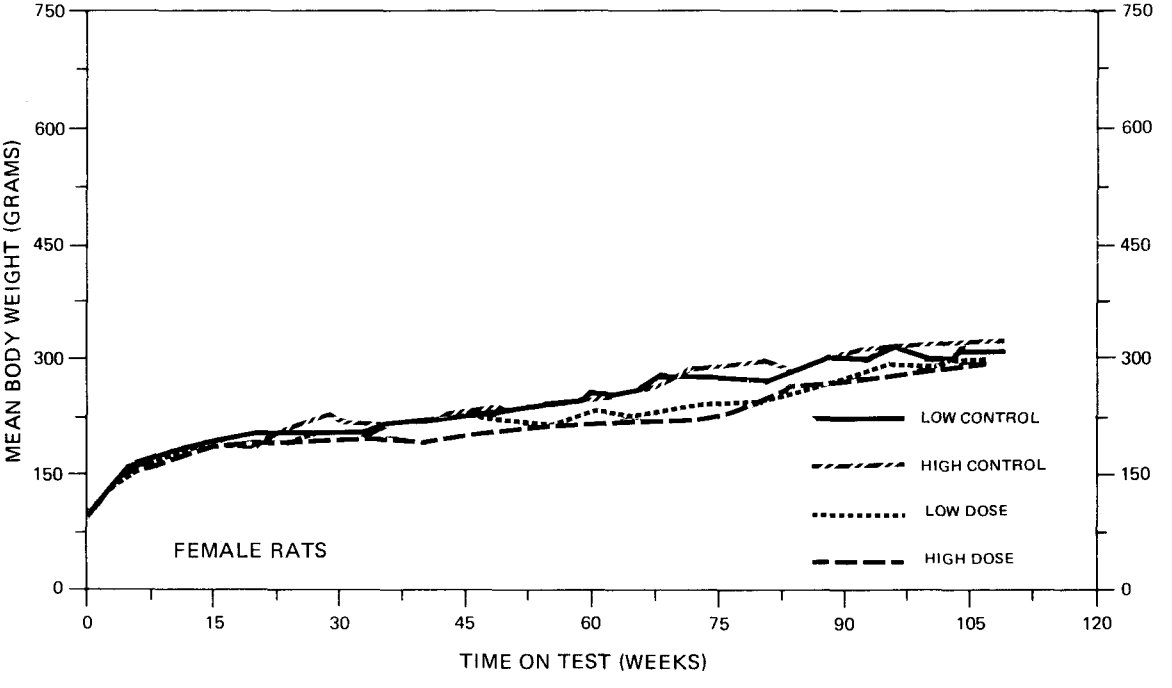
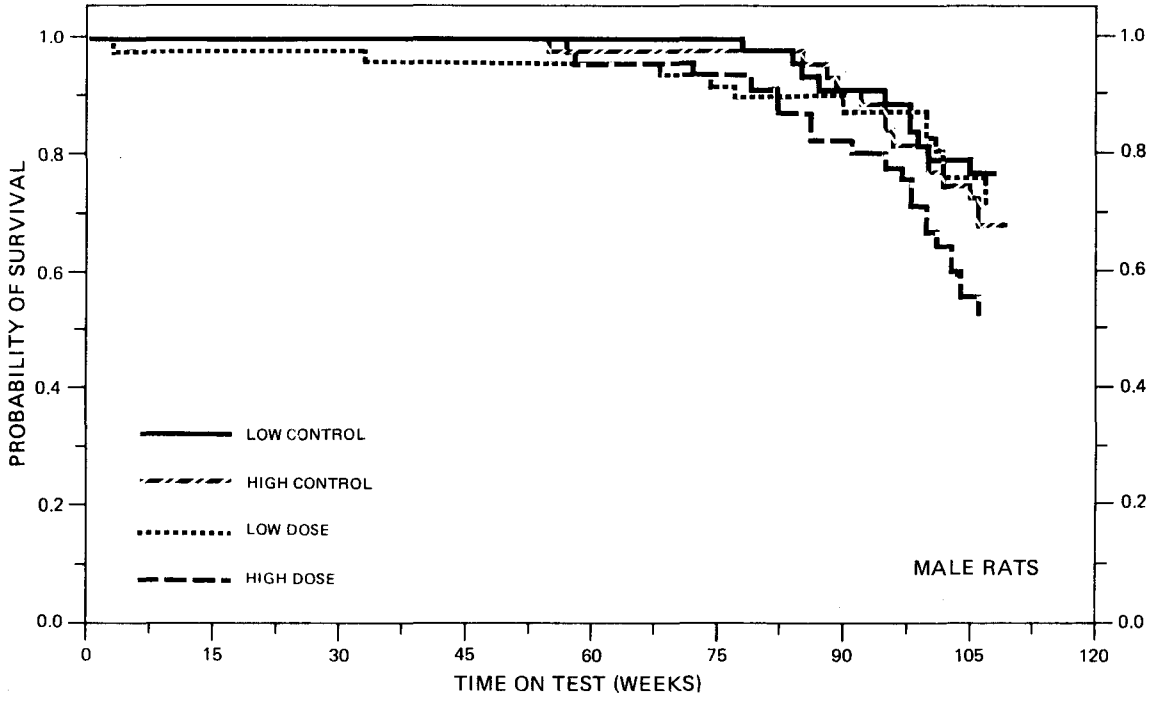


FIGURE 2
GROWTH CURVES FOR HYDRAZOBENZENE CHRONIC STUDY RATS



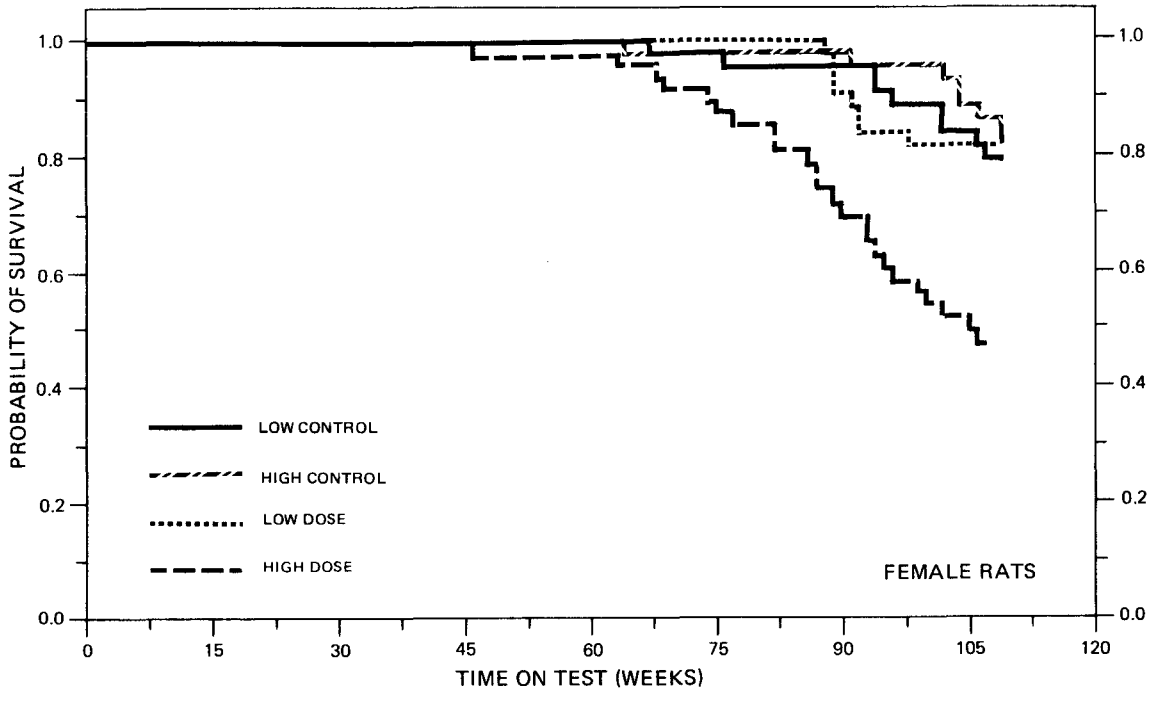


FIGURE 3
SURVIVAL COMPARISONS OF HYDRAZOBENZENE CHRONIC STUDY RATS

dose, 78 percent (39/50) of the low dose, 73 percent (36/49) of the high dose controls and 70 percent (35/50) of the low dose controls were still alive. There were adquate numbers of male rats at risk from late-developing tumors.

For female rats the Cox test indicated that the mortality was significantly ($P < 0.001$) greater in the high dose group than in the high dose control. For both the treated and untreated female rats, five animals from each group were sacrificed in week 78. There were adequate numbers of female rats at risk from late-developing tumors with 50 percent (25/50) of the high dose, 74 percent (37/50) of the low dose, 86 percent (43/50) of the high dose control and 78 percent (39/50) of the low dose control surviving in week 100.

C. Pathology

Histopathologic findings on neoplasms in rats are summarized in Appendix A (Tables A1 and A2); findings on nonneoplastic lesions are summarized in Appendix C (Tables C1 and C2).

The incidence of neoplasms in most organs in the treated rats was comparable to that of the control groups. However, hydrazobenzene feeding increased the development of neoplasms of the liver, auditory sebaceous gland, and mammary gland (females).

Hepatocellular carcinomas were found in 36 treated male rats as early as week 72 (5/49 [10 percent] low dose and 31/49 [63 percent] high dose), in none of the treated female rats, and in 1/95 (1 percent) control male rats. Neoplastic nodules found in treated males

and females and in control males involved a few lobules, were well demarcated, and compressed the adjacent normal liver cells in some areas. Cells in these nodules were large, and cytoplasm either acidophilic or basophilic. Nuclei were vesicular. The hepatocellular carcinomas involved one or many lobes of the liver. The tumors appeared well-differentiated. Lobular architecture was distorted within the neoplasms, and the liver plates were several cells thick. In some tumors, the cells were arranged in a glandular pattern, and there appeared to be a slight pleomorphism in size of transformed hepatocytes. The cytoplasm of a majority of cells was acidophilic; in some it was vacuolated. Nuclei were large and vesicular, and nucleoli prominent. Both normal and abnormal mitotic figures were present. Sinusoids were distended and engorged with blood, and in some there were a few hematopoietic cells. Small areas of necrosis, cellular debris, and inflammatory cells were observed. Hepatocellular carcinomas metastasized to the lung in six treated male rats.

Neoplasms of the Zymbal's gland or ear canal occurred in 1/95 (1 percent) control male rats, 2/50 (4 percent) low dose males, 7/49 (14 percent) high dose males, 0/98 control females, 1/50 (2 percent) low dose females, and 2/50 (4 percent) high dose females. The tumors in eight of the treated rats were considered to be squamous-cell carcinomas, one was a squamous-cell papilloma (high dose male), and the remaining two were sebaceous adenocarcinomas. A squamous-cell carcinoma in one low dose male rat had metastasized to the lung.

Adenocarcinomas of the mammary gland were found in 1/48 (2 percent) low dose control females and in nine treated females (3/50 [6 percent] low dose and 6/50 [12 percent] high dose). This tumor had metastasized to the lung in one high dose female. The incidence of fibroadenoma of the mammary gland in treated female rats was lower than in female controls.

Neoplasms of the clitoral gland in treated females and the preputial gland in treated males as well as adrenal pheochromocytomas in high dose males appeared to be slightly increased when compared with control animals.

There was a variety of nonneoplastic lesions observed, but none appeared to be compound-related.

Based upon this histopathologic examination evidence was provided for the carcinogenicity of hydrazobenzene in Fischer 344 rats as dietary administration of the compound was associated with a dose-related increase in the incidence of hepatic neoplasms and auditory sebaceous gland neoplasms and a moderately increased incidence of mammary gland adenocarcinomas.

D. Statistical Analyses of Results

The results of the statistical analyses of tumor incidence in rats are summarized in Tables 3 and 4. The analysis is included for every type of tumor in either sex where at least two such tumors were observed in at least one of the control or hydrazobenzene-dosed groups

TABLE 3

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT
SPECIFIC SITES IN MALE RATS TREATED WITH HYDRAZOBENZENE[a]

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Skin excluding Skin of Ear: Squamous-Cell Papilloma[b]	1/47(0.02)	0/48(0.00)	1/50(0.02)	3/49(0.06)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.940	Infinite
Lower Limit	---	---	0.012	0.590
Upper Limit	---	---	72.331	Infinite
Weeks to First Observed Tumor	108	---	90	106
Subcutaneous Tissue: Fibroma[b]	7/47(0.15)	3/48(0.06)	2/50(0.04)	1/49(0.02)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.269	0.327
Lower Limit	---	---	0.028	0.006
Upper Limit	---	---	1.325	3.898
Weeks to First Observed Tumor	98	95	90	106
Lung: Alveolar/Bronchiolar Adenoma or Alveolar/Bronchiolar Carcinoma[b]	1/47(0.02)	1/48(0.02)	3/49(0.06)	1/48(0.02)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	2.878	1.000
Lower Limit	---	---	0.241	0.013
Upper Limit	---	---	147.900	6.886
Weeks to First Observed Tumor	108	109	107	106

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Hematopoietic System: Leukemia or Malignant Lymphoma[b]	5/47(0.11)	6/48(0.13)	2/50(0.04)	1/49(0.02)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.376	0.163
Lower Limit	---	---	0.037	0.004
Upper Limit	---	---	2.172	1.274
Weeks to First Observed Tumor	84	92	102	106
Spleen: Hemangiosarcoma[b]	0/47(0.00)	0/48(0.00)	0/49(0.00)	3/49(0.06)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	---	Infinite
Lower Limit	---	---	---	0.590
Upper Limit	---	---	---	Infinite
Weeks to First Observed Tumor	---	---	---	86
Liver: Hepatocellular Carcinoma[b]	0/47(0.00)	1/48(0.02)	5/49(0.10)	31/49(0.63)
P Values[c]	---	---	P = 0.031	P < 0.001
Relative Risk (Control)[d]	---	---	Infinite	30.367
Lower Limit	---	---	1.212	5.548
Upper Limit	---	---	Infinite	1170.322
Weeks to First Observed Tumor	---	109	107	72

TABLE 3 (Continued)

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Liver: Neoplastic Nodule or Hepatocellular Carcinoma[b]	5/47(0.11)	1/48(0.02)	13/49(0.27)	37/49(0.76)
P Values[c]	---	---	P = 0.040	P < 0.001
Relative Risk (Control)[d]	---	---	2.494	38.204
Lower Limit	---	---	0.914	7.290
Upper Limit	---	---	8.254	1395.851
Weeks to First Observed Tumor	108	109	102	72
Adrenal: Pheochromocytoma or Pheochromocytoma, Malignant[b]	7/47(0.15)	8/47(0.17)	7/48(0.15)	16/46(0.35)
P Values[c]	---	---	N.S.	P = 0.042
Relative Risk (Control)[d]	---	---	0.979	2.043
Lower Limit	---	---	0.318	0.922
Upper Limit	---	---	3.019	4.939
Weeks to First Observed Tumor	108	106	74	78
Pituitary: Adenoma NOS or Chromophobe Adenoma[b]	8/47(0.17)	9/38(0.24)	8/46(0.17)	2/29(0.07)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	1.022	0.291
Lower Limit	---	---	0.365	0.033
Upper Limit	---	---	2.856	1.267
Weeks to First Observed Tumor	108	85	78	106

TABLE 3 (Continued)

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Thyroid: C-Cell Carcinoma[b]	2/46(0.04)	1/48(0.02)	2/41(0.05)	2/44(0.05)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	1.122	2.182
Lower Limit	---	---	0.085	0.118
Upper Limit	---	---	14.845	125.735
Weeks to First Observed Tumor	108	109	102	82
Thyroid: C-Cell Adenoma or C-Cell Carcinoma[b]	4/46(0.09)	1/48(0.02)	2/41(0.05)	4/44(0.09)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.561	4.364
Lower Limit	---	---	0.053	0.454
Upper Limit	---	---	3.687	209.675
Weeks to First Observed Tumor	108	109	102	82
Pancreatic Islets: Islet-Cell Adenoma or Islet-Cell Carcinoma[b]	4/45(0.09)	0/46(0.00)	4/47(0.09)	1/45(0.02)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.957	Infinite
Lower Limit	---	---	0.189	0.055
Upper Limit	---	---	4.845	Infinite
Weeks to First Observed Tumor	85	---	107	106

TABLE 3 (Continued)

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Testis: Interstitial-Cell Tumor[b]	44/47(0.94)	42/47(0.89)	46/49(0.94)	43/48(0.90)
P Values[c]	---	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	---	1.003	1.002
Lower Limit	---	---	0.907	0.869
Upper Limit	---	---	1.109	1.156
Weeks to First Observed Tumor	78	78	74	78
Zymbal's Gland: Squamous-Cell Carcinoma[b]	0/47(0.00)	0/48(0.00)	1/50(0.02)	5/49(0.10)
P Values[c]	---	---	N.S.	P = 0.030
Relative Risk (Control)[d]	---	---	Infinite	Infinite
Lower Limit	---	---	0.050	1.237
Upper Limit	---	---	Infinite	Infinite
Weeks to First Observed Tumor	---	---	107	86
Ear Canal, Zymbal's Gland, and Skin of the Ear: Squamous-Cell Carcinoma or Squamous-Cell Papilloma[b]	1/47(0.02)	0/48(0.00)	2/50(0.04)	7/49(0.14)
P Values[c]	---	---	N.S.	P = 0.007
Relative Risk (Control)[d]	---	---	1.880	Infinite
Lower Limit	---	---	0.101	1.904
Upper Limit	---	---	108.696	Infinite
Weeks to First Observed Tumor	87	---	68	78

Table 3 (Concluded)

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Body Cavities: Mesothelioma NOS[b]	2/47(0.04)	2/48(0.04)	3/50(0.06)	5/49(0.10)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	1.440	2.449
Lower Limit	---	---	0.173	0.416
Upper Limit	---	---	16.632	24.226
Weeks to First Observed Tumor	95	106	100	78

[a]Treated groups received time-weighted average doses of 0.008 or 0.03 percent in feed.

[b]Number of tumor-bearing animals/number of animals examined at site (proportion).

[c]The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. A negative designation (N) indicates a lower incidence in the treated group than in the control group.

[d]The 95% confidence interval of the relative risk of the treated group to the control group.

TABLE 4

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT SPECIFIC SITES IN FEMALE RATS TREATED WITH HYDRAZOBENZENE[a]

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Hematopoietic System: Leukemia[b]	3/48(0.06)	5/50(0.10)	0/50(0.00)	0/50(0.00)
P Values[c]	---	---	P = 0.028(N)	P = 0.028(N)
Relative Risk (Control)[d]	---	---	0.000	0.000
Lower Limit	---	---	0.000	0.000
Upper Limit	---	---	1.596	10.793
Weeks to First Observed Tumor	107	104	---	---
Liver: Neoplastic Nodule[b]	0/47(0.00)	0/50(0.00)	0/50(0.00)	6/50(0.12)
P Values[c]	---	---	N.S.	P = 0.013
Relative Risk (Control)[d]	---	---	---	Infinite
Lower Limit	---	---	---	1.560
Upper Limit	---	---	---	Infinite
Weeks to First Observed Tumor	---	---	---	77
Pituitary: Adenoma NOS or Chromophobe Adenoma[b]	18/46(0.39)	17/40(0.43)	21/47(0.45)	13/37(0.35)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	1.142	0.827
Lower Limit	---	---	0.675	0.434
Upper Limit	---	---	1.946	1.538
Weeks to First Observed Tumor	94	78	78	69

TABLE 4 (Continued)

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Adrenal: Pheochromocytoma or Pheochromocytoma, Malignant[b]	1/47(0.02)	3/49(0.06)	3/49(0.06)	3/49(0.06)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	2.878	1.000
Lower Limit	---	---	0.241	0.140
Upper Limit	---	---	147.907	7.126
Weeks to First Observed Tumor	108	109	107	95
Thyroid: C-Cell Carcinoma[b]	3/46(0.07)	1/45(0.02)	3/50(0.06)	3/46(0.07)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.920	2.935
Lower Limit	---	---	0.129	0.247
Upper Limit	---	---	6.556	0.618
Weeks to First Observed Tumor	109	109	107	94
Thyroid: C-Cell Adenoma or C-Cell Carcinoma[b]	3/46(0.07)	2/45(0.05)	5/50(0.10)	3/46(0.07)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	1.533	1.467
Lower Limit	---	---	0.317	0.176
Upper Limit	---	---	9.398	6.894
Weeks to First Observed Tumor	109	109	107	94

TABLE 4 (Continued)

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Mammary Gland: Adenocarcinoma NOS[b]	1/48(0.02)	0/50(0.00)	3/50(0.06)	6/50(0.12)
P Values[c]	---	---	N.S.	P = 0.013
Relative Risk (Control)[d]	---	---	2.880	Infinite
Lower Limit	---	---	0.241	1.600
Upper Limit	---	---	148.100	Infinite
Weeks to First Observed Tumor	109	---	92	87
Mammary Gland: Fibroadenoma[b]	14/48(0.29)	19/50(0.38)	9/50(0.18)	9/50(0.18)
P Values[c]	---	---	N.S.	P = 0.022(N)
Relative Risk (Control)[d]	---	---	0.617	0.474
Lower Limit	---	---	0.261	0.211
Upper Limit	---	---	1.381	0.983
Weeks to First Observed Tumor	76	106	107	46
Uterus: Endometrial Stromal Polyp[b]	15/47(0.32)	10/50(0.20)	10/50(0.20)	5/48(0.10)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.627	0.521
Lower Limit	---	---	0.282	0.150
Upper Limit	---	---	1.337	1.540
Weeks to First Observed Tumor	78	78	78	78

TABLE 4 (Continued)

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Uterus: Endometrial Stromal Sarcoma[b]	0/47(0.00)	1/50(0.02)	3/50(0.06)	1/48(0.02)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	Infinite	1.042
Lower Limit	---	---	0.566	0.014
Upper Limit	---	---	Infinite	80.093
Weeks to First Observed Tumor	---	104	88	82
Uterus and Uterus Endometrium: Carcinoma NOS or Adenocarcinoma NOS[b]	2/47(0.04)	1/50(0.02)	5/50(0.10)	3/48(0.06)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	2.350	3.125
Lower Limit	---	---	0.407	0.262
Upper Limit	---	---	23.761	160.536
Weeks to First Observed Tumor	109	109	89	106
Clitoral Gland: Adenoma NOS[b]	0/48(0.00)	2/50(0.04)	0/50(0.00)	6/50(0.12)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	---	3.000
Lower Limit	---	---	---	0.569
Upper Limit	---	---	---	29.250
Weeks to First Observed Tumor	---	104	---	68

TABLE 4 (Concluded)

[a]Treated groups received time-weighted average doses of 0.004 or 0.01 percent in feed.

[b]Number of tumor-bearing animals/number of animals examined at site (proportion).

[c]The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. A negative designation (N) indicates a lower incidence in the treated group than in the control group.

[d]The 95% confidence interval of the relative risk of the treated group to the control group.

and where such tumors were observed in at least 5 percent of the group. Cochran-Armitage tests were not used in these analyses.

High incidences of hepatocellular carcinomas in male rats and neoplastic nodules of the liver in rats of both sexes were observed. For males the Fisher exact test indicated a significantly elevated incidence of hepatocellular carcinomas in the high dose ($P < 0.001$) compared to its controls. The Fisher exact test comparing the low dose male rats with their controls had a significance level of $P = 0.031$, a marginal result which was not significant under the Bonferroni criterion. For high dose female rats, the Fisher exact test was significant ($P = 0.013$) when the high dose incidence was compared to that of the high dose control. Based upon these statistical results, the administration of hydrazobenzene was associated with an elevated incidence of neoplastic nodules in female rats and hepatocellular carcinomas in male rats.

In male rats an elevated combined incidence of squamous-cell carcinomas or squamous-cell papillomas of the Zymbal's gland, the ear canal, or the skin of the ear was observed. The Fisher exact test comparing the incidence of these tumors in the high dose to that in the high dose control was significant ($P = 0.007$). Based upon these statistical results, administration was associated with an elevated incidence of squamous-cell neoplasms in male rats.

The elevated incidence of adrenal pheochromocytomas in the high dose male rats relative to the high dose controls yielded a

significance level of P = 0.042 for the Fisher exact test, a marginal result which was not significant under the Bonferroni criterion.

In female rats a high incidence of mammary gland adenocarcinomas was observed. The Fisher exact test indicated a significantly (P = 0.013) elevated incidence of these tumors in the high dose group relative to that in the high dose controls. The historical incidence of mammary adenocarcinomas in female Fischer 344 rats observed at Mason Research Institute for the NCI Carcinogenesis Testing Program was 8/585 (1 percent). Based upon these statistical results the administration of hydrazobenzene was associated with an elevated incidence of mammary gland adenocarcinomas in female rats.

Negative associations resulted from the Fisher exact test comparing the incidence of leukemia or malignant lymphoma in both low and high dose female rats to the incidence in their respective controls. These results appear to be spurious.

In female rats negative results were obtained from the Fisher exact test comparison of the high dose rats to the high dose controls for the incidence of mammary gland fibroadenomas. The incidence of this tumor in the high dose controls (19/50 or 38 percent) was high compared to the 115/585 (20 percent) observed in the Fischer 344 historical untreated female control rats maintained at Mason Research Institute for the NCI Carcinogenesis Testing Program.

In summary, the administration of hydrazobenzene was associated with increased incidences of hepatocellular carcinomas and of

squamous-cell neoplasms of the Zymbal's gland, ear canal, and skin of ear in male rats and with increased incidences of neoplastic nodules of the liver and of mammary adenocarcinomas in female rats.

IV. CHRONIC TESTING RESULTS: MICE

A. Body Weights and Clinical Observations

No distinct pattern of mean body weight depression was evident for low dose mice of either sex. Depression of mean group body weight relative to controls was observed, however, for male and female high dose mice after week 28 (Figure 4).

No clinical abnormalities were recorded for treated or control mice of either sex.

B. Survival

The estimated probabilities of survival for male and female mice in the control and hydrazobenzene-dosed groups are shown in Figure 5.

For male mice the Cox test indicated a significantly greater mortality in the high dose group than in the high dose control. Five male mice were sacrificed from the high dose group and from each control group in week 78. In addition, five mice from the high dose male control group were sacrificed in week 49. There were adequate numbers of male mice at risk from late-developing tumors with 66 percent (33/50) of the high dose, 88 percent (44/50) of the low dose, 78 percent (39/50) of the high dose control, and 86 percent (43/50) of the low dose control surviving to the termination of the study.

For female mice the Cox test indicated a significantly greater mortality in the high dose group than in the high dose control. As with the male mice, five high dose females and five females from

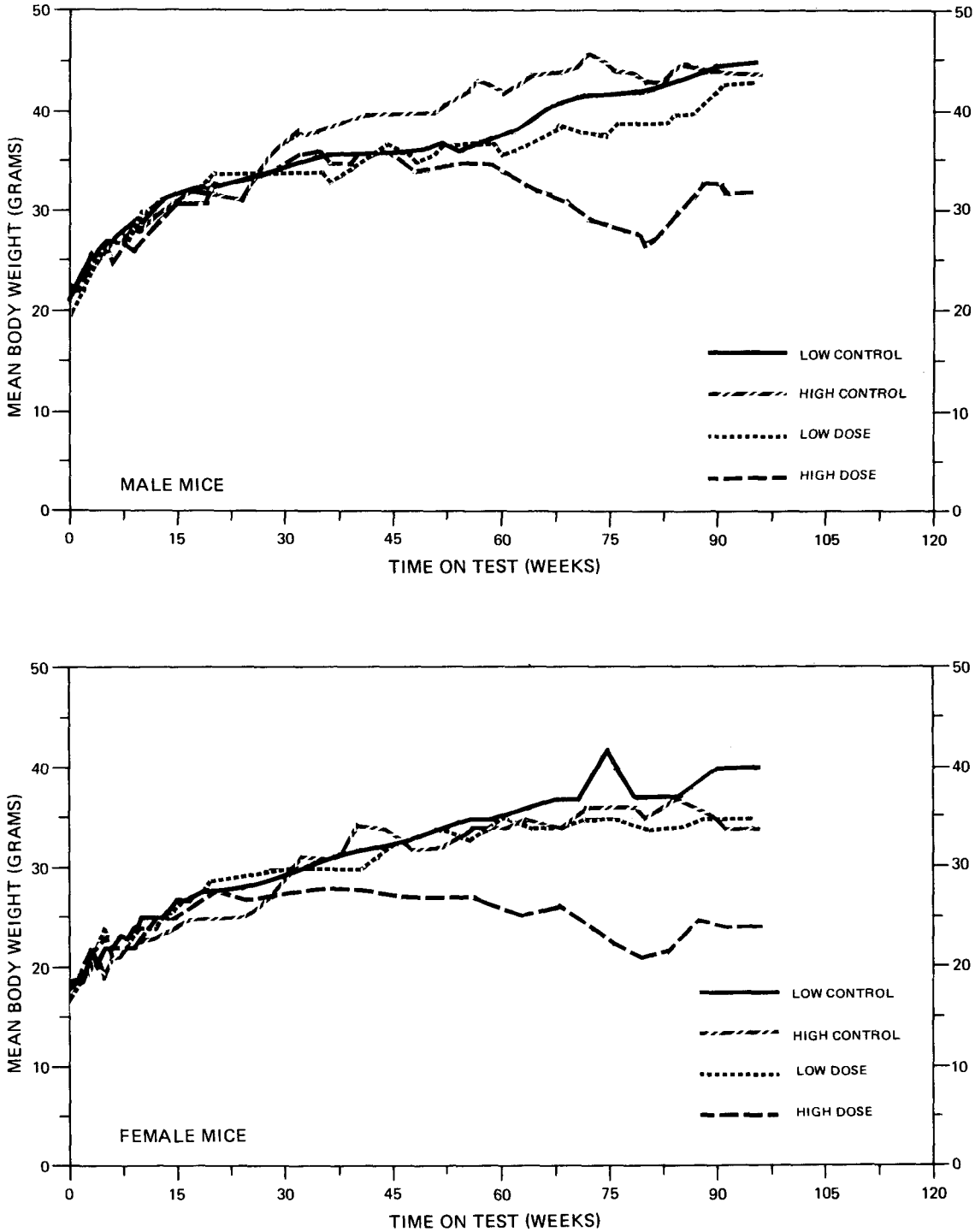
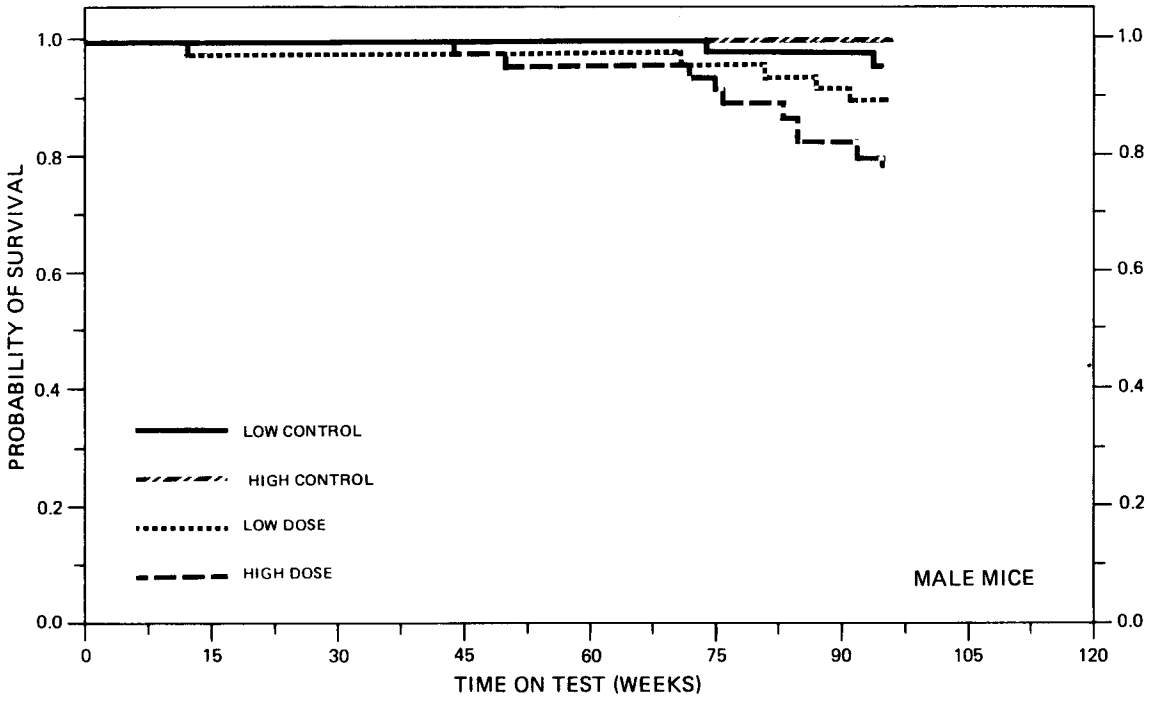


FIGURE 4
GROWTH CURVES FOR HYDRAZOBENZENE CHRONIC STUDY MICE



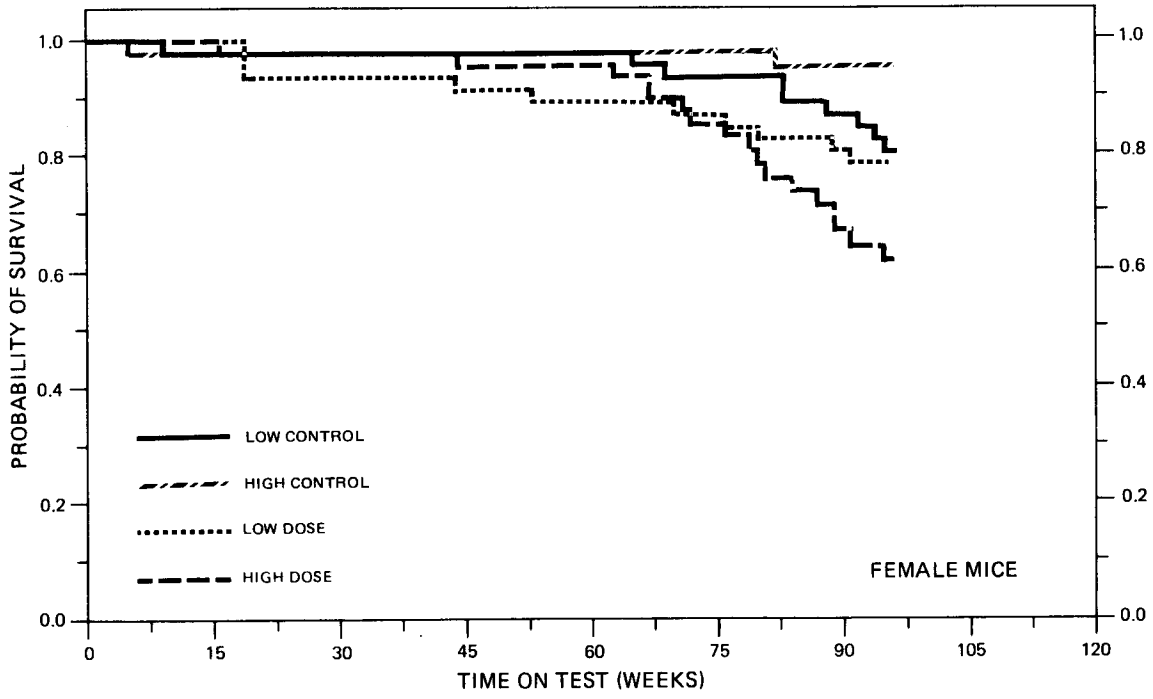


FIGURE 5
SURVIVAL COMPARISONS OF HYDRAZOBENZENE CHRONIC STUDY MICE

each of the control groups were sacrificed in week 78 with an additional five from the high dose control being sacrificed in week 49. Survival of the female mice was relatively good with 52 percent (26/50) of the high dose, 79 percent (37/47) of the low dose, 76 percent (38/50) of the high dose control, and 72 percent (36/50) of the low dose control animals living until the termination of the study. Thus, there were adequate numbers of female mice at risk from late-developing tumors.

C. Pathology

Histopathologic findings on neoplasms in mice are summarized in Appendix B (Tables B1 and B2); findings on nonneoplastic lesions are summarized in Appendix D (Tables D1 and D2).

There was an increased incidence of hepatocellular carcinomas in female mice (20/43 [47 percent] high dose, 4/39 [10 percent] low dose, and 3/97 [3 percent] controls). Hepatic neoplasms in these mice involved a part or a whole lobe and compressed the surrounding normal hepatic parenchyma.

The cytoplasm of the neoplastic hepatocytes varied in tinctorial quality. It was usually basophilic or acidophilic and was occasionally vacuolated. Nuclei were large and vesicular, and mitotic figures were infrequently observed. In some cells, there were nuclear inclusions. Sinusoids and blood vessels in the tumors were dilated. Hepatocellular carcinomas metastasized to the lung in one low dose male, two control males, and one control female.

These mice had a variety of other neoplastic and nonneoplastic lesions, but none appeared to be compound-related.

Based upon this histopathologic examination hydrazobenzene is carcinogenic to female B6C3F1 mice as there was a dose-related increase in the incidence of hepatocellular carcinomas. The compound does not appear to be carcinogenic to male mice.

D. Statistical Analyses of Results

The results of the statistical analyses of tumor incidence in mice are summarized in Tables 5 and 6. The analysis is included for every type of tumor in either sex where at least two such tumors were observed in at least one of the control or hydrazobenzene-dosed groups and where such tumors were observed in at least 5 percent of the group. Cochran-Armitage tests were not used in these analyses since the low dose groups and their controls were started at different times from the high dose groups and their controls.

Female mice in the treated groups showed increased incidences of hepatocellular carcinoma. The Fisher exact test comparing the incidence in the high dose group with that in the high dose control was significant ($P < 0.001$). The Fisher exact test comparing the high dose to the high dose control for the increased incidence of hepatocellular adenoma or hepatocellular carcinoma was also significant ($P < 0.001$).

TABLE 5

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT SPECIFIC SITES IN MALE MICE TREATED WITH HYDRAZOBENZENE[a]

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Lung: Alveolar/Bronchiolar Carcinoma[b]	5/50(0.10)	5/49(0.10)	1/47(0.02)	0/46(0.00)
P Values[c]	---	---	N.S.	P = 0.033(N)
Relative Risk (Control)[d]	---	---	0.213	0.000
Lower Limit	---	---	0.005	0.000
Upper Limit	---	---	1.804	0.843
Weeks to First Observed Tumor	95	96	95	---
Lung: Alveolar/Bronchiolar Adenoma or Alveolar/Bronchiolar Carcinoma[b]	5/50(0.10)	10/49(0.20)	2/47(0.04)	3/46(0.07)
P Values[c]	---	---	N.S.	P = 0.046(N)
Relative Risk (Control)[d]	---	---	0.426	0.320
Lower Limit	---	---	0.042	0.060
Upper Limit	---	---	2.454	1.149
Weeks to First Observed Tumor	95	96	95	95
Hematopoietic System: Malignant Lymphoma[b]	5/50(0.10)	5/49(0.10)	4/49(0.08)	1/48(0.02)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.816	0.204
Lower Limit	---	---	0.171	0.004
Upper Limit	---	---	3.567	1.732
Weeks to First Observed Tumor	74	96	95	95

TABLE 5 (Continued)

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Hematopoietic System: Malignant Lymphoma or Leukemia[b]	5/50(0.10)	5/49(0.10)	4/49(0.08)	3/48(0.06)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.816	0.613
Lower Limit	---	---	0.171	0.100
Upper Limit	---	---	3.567	2.965
Weeks to First Observed Tumor	74	96	95	85
Liver: Hepatocellular Carcinoma[b]	12/50(0.24)	6/48(0.13)	11/47(0.23)	8/46(0.17)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	0.975	1.391
Lower Limit	---	---	0.433	0.460
Upper Limit	---	---	2.169	4.492
Weeks to First Observed Tumor	94	78	87	78
Adrenal: Pheochromocytoma[b]	0/49(0.00)	1/44(0.02)	1/45(0.02)	3/42(0.07)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	Infinite	3.143
Lower Limit	---	---	0.058	0.265
Upper Limit	---	---	Infinite	0.893
Weeks to First Observed Tumor	---	96	95	92

TABLE 5 (Concluded)

[a]Treated groups received time-weighted average doses of 0.008 or 0.04 percent in feed.

[b]Number of tumor-bearing animals/number of animals examined at site (proportion).

[c]The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. A negative designation (N) indicates a lower incidence in the treated group than in the control group.

[d]The 95% confidence interval of the relative risk of the treated group to the control group.

TABLE 6

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT SPECIFIC SITES IN FEMALE MICE TREATED WITH HYDRAZOBENZENE[a]

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Hematopoietic System: Malignant Lymphoma[b]	5/48(0.10)	2/50(0.04)	10/40(0.28)	5/44(0.11)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	2.400	2.841
Lower Limit	---	---	0.818	0.492
Upper Limit	---	---	8.189	8.601
Weeks to First Observed Tumor	96	96	76	67
Hematopoietic System: Malignant Lymphoma or Leukemia[b]	7/48(0.15)	2/50(0.04)	10/40(0.28)	6/44(0.14)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	1.714	3.341
Lower Limit	---	---	0.649	0.635
Upper Limit	---	---	4.795	32.410
Weeks to First Observed Tumor	83	96	76	67
Lung: Alveolar/Bronchiolar Adenoma or Alveolar/Bronchiolar Carcinoma[b]	2/46(0.04)	3/50(0.06)	3/38(0.08)	2/40(0.05)
P Values[c]	---	---	N.S.	N.S.
Relative Risk (Control)[d]	---	---	1.816	0.833
Lower Limit	---	---	0.219	0.072
Upper Limit	---	---	20.760	6.902
Weeks to First Observed Tumor	96	78	95	95

TABLE 6 (Concluded)

TOPOGRAPHY:MORPHOLOGY	LOW DOSE CONTROL	HIGH DOSE CONTROL	LOW DOSE	HIGH DOSE
Liver: Hepatocellular Carcinoma[b]	2/47(0.04)	1/50(0.02)	4/39(0.10)	20/43(0.47)
P Values[c]	---	---	N.S.	P < 0.001
Relative Risk (Control)[d]	---	---	2.410	23.260
Lower Limit	---	---	0.366	4.027
Upper Limit	---	---	25.430	922.300
Weeks to First Observed Tumor	94	96	95	78
Liver: Hepatocellular Adenoma or Hepatocellular Carcinoma[b]	2/47(0.04)	1/50(0.02)	4/39(0.10)	22/43(0.51)
P Values[c]	---	---	N.S.	P < 0.001
Relative Risk (Control)[d]	---	---	2.410	25.581
Lower Limit	---	---	0.366	4.493
Upper Limit	---	---	25.434	1006.419
Weeks to First Observed Tumor	94	96	95	78

[a]Treated groups received time-weighted average doses of 0.004 or 0.04 percent in feed.

[b]Number of tumor-bearing animals/number of animals examined at site (proportion).

[c]The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when P < 0.05; otherwise, not significant (N.S.) is indicated. A negative designation (N) indicates a lower incidence in the treated group than in the control group.

[d]The 95% confidence interval of the relative risk of the treated group to the control group.

Based upon these statistical results the administration of hydrazobenzene was associated with an elevated incidence of hepatocellular carcinoma in female mice.

For male mice the Fisher exact test comparing the high dose animals to the high dose control for alveolar/bronchiolar carcinoma was not significant under the Bonferroni criterion. Similarly, when incidences of both alveolar/bronchiolar adenoma and alveolar/bronchiolar carcinoma were combined the results of the Fisher exact test were not significant under the Bonferroni criterion.

V. DISCUSSION

Adequate numbers of animals in all groups of rats and mice in this study survived sufficiently long to be at risk from late-developing tumors.

The methods used for chemical analyses were not quantitative, and, therefore, did not indicate the purity of the compound. No qualitative analyses for identification of impurities were performed.

The liver was a target organ for carcinogenicity in the rat. The incidence of hepatocellular carcinomas was significantly increased in dosed male rats and the incidence of neoplastic nodules of the liver was significantly increased in dosed female rats. The combined incidence of squamous-cell carcinomas and squamous-cell papillomas of the Zymbal's gland, ear canal or skin of the ear was statistically significant in male rats. The incidence of mammary gland adenocarcinoma was significantly increased for female rats.

The liver was the target organ for carcinogenicity in female mice. The incidence of hepatocellular carcinoma in the high dose female mice was significant. No significantly increased incidences of liver tumors were, however, observed among dosed male mice when compared to controls.

The evidence for carcinogenicity of hydrazobenzene in this bioassay is supported by positive results for carcinogenicity reported in other studies. Hydrazobenzene was found to be carcinogenic in both rats and mice following exposure via subcutaneous injection

(Pliss, 1974). Tumors of the uterus, mammary gland, concha cimbae (Zymbal's gland), and liver were noted in rats while subcutaneous sarcomas and hepatic tumors were noted in mice (Pliss, 1974). Concurrent subcutaneous administration of benzidine sulfate and hydrazobenzene to rats resulted in an increased incidence of bladder cancer and a decreased latent period when compared to the tumor incidences and latent periods noted following administration of the individual compounds (Kurlyandskii et al., 1976).

Under the conditions of this bioassay, hydrazobenzene was carcinogenic to Fischer 344 rats of both sexes, causing increased incidences of hepatocellular carcinoma and Zymbal's gland squamous-cell neoplasms in male rats, neoplastic nodules of the liver in female rats, and mammary adenocarcinomas in female rats. Hydrazobenzene was also carcinogenic to female B6C3F1 mice, causing an increased incidence of hepatocellular carcinomas. The compound was not carcinogenic to male B6C3F1 mice.

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APPENDIX A

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN RATS TREATED WITH HYDRAZOBENZENE

TABLE A1
SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE RATS TREATED WITH HYDRAZOBENZENE

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
ANIMALS INITIALLY IN STUDY	50	49	50	50
ANIMALS MISSING	2			
ANIMALS NECROPSIED	47	48	50	49
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	47	48	49	49
INTEGUMENTARY SYSTEM				
*SKIN	(47)	(48)	(50)	(49)
SQUAMOUS CELL PAPILLOMA	1 (2%)		1 (2%)	4 (8%)
BASAL-CELL CARCINOMA				1 (2%)
LEIOMYOSARCOMA	1 (2%)			
*SUBCUT TISSUE	(47)	(48)	(50)	(49)
UNDIFFERENTIATED CARCINOMA			1 (2%)	
BASAL-CELL CARCINOMA	1 (2%)			
SARCOMA, NOS		1 (2%)		1 (2%)
FIBROMA	7 (15%)	3 (6%)	2 (4%)	1 (2%)
FIBROSARCOMA		1 (2%)		
MYOEPITHELIOMA			1 (2%)	
RESPIRATORY SYSTEM				
#LUNG	(47)	(48)	(49)	(48)
SQUAMOUS CELL CARCINOMA				1 (2%)
SQUAMOUS CELL CARCINOMA, METASTA			1 (2%)	
HEPATOCELLULAR CARCINOMA, METAST			1 (2%)	5 (10%)
ALVEOLAR/BRONCHIOLAR ADENOMA	1 (2%)		2 (4%)	
ALVEOLAR/BRONCHIOLAR CARCINOMA		1 (2%)	1 (2%)	1 (2%)
PHEOCHROMOCYTOMA, METASTATIC		1 (2%)		
SARCOMA, NOS, METASTATIC				1 (2%)
HEMATOPOIETIC SYSTEM				
*MULTIPLE ORGANS	(47)	(48)	(50)	(49)
MALIGNANT LYMPHOMA, NOS	1 (2%)	1 (2%)		
LEUKEMIA,NOS	1 (2%)	1 (2%)		
UNDIFFERENTIATED LEUKEMIA	1 (2%)			
MYELOMONOCYTIC LEUKEMIA	1 (2%)	4 (8%)	1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE A1 (CONTINUED)

	LOW DOSE CONTROL(UNTP) 01-0055	HIGH DOSE CONTROL(UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
#SPLEEN	(47)	(48)	(49)	(48)
MUCINOUS ADENOCARCINOMA, METASTA	1 (2%)			
HEMANGIOSARCOMA				3 (6%)
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE	1 (2%)			
MALIG.LYMPHOMA, HISTIOCYTIC TYPE				1 (2%)
#MEDIASTINAL L.NODE	(42)	(44)	(48)	(27)
MUCINOUS ADENOCARCINOMA, METASTA	1 (2%)			
*CARDIOVASCULAR SYSTE	(47)	(48)	(50)	(49)
UNDIFFERENTIATED LEUKEMIA			1 (2%)	
#LIVER	(47)	(48)	(49)	(49)
KUPFFER-CELL SARCOMA			1 (2%)	
#THYMUS	(42)	(23)	(31)	(20)
THYMOMA				1 (5%)
CIRCULATORY SYSTEM				
#HEART	(47)	(48)	(49)	(47)
SQUAMOUS CELL CARCINOMA, METASTA				1 (2%)
DIGESTIVE SYSTEM				
*ORAL CAVITY	(47)	(48)	(50)	(49)
FIBROSARCOMA	1 (2%)			
#SALIVARY GLAND	(46)	(47)	(46)	(46)
ADENOCARCINOMA, NOS		1 (2%)		
SARCOMA, NOS		1 (2%)		
#LIVER	(47)	(48)	(49)	(49)
NEOPLASTIC NODULE	5 (11%)		8 (16%)	6 (12%)
HEPATOCELLULAR CARCINOMA		1 (2%)	5 (10%)	31 (63%)
#PANCREAS	(45)	(46)	(47)	(45)
MUCINOUS ADENOCARCINOMA, METASTA	1 (2%)			
ACINAR-CELL ADENOMA				1 (2%)
#STOMACH	(47)	(48)	(49)	(47)
SQUAMOUS CELL PAPILLOMA				2 (4%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
MUCINOUS ADENOCARCINOMA, METASTA	1 (2%)			
#SMALL INTESTINE	(47)	(46)	(48)	(39)
LEIOMYOSARCOMA				1 (3%)
#ILEUM	(47)	(46)	(48)	(39)
SARCOMA, NOS		1 (2%)		
#COLON	(46)	(46)	(47)	(33)
MUCINOUS ADENOCARCINOMA	1 (2%)			
URINARY SYSTEM				
#KIDNEY	(47)	(48)	(49)	(48)
LIPOMA	1 (2%)			
ENDOCRINE SYSTEM				
#PITUITARY	(47)	(38)	(46)	(29)
ADENOMA, NOS	8 (17%)	9 (24%)	6 (13%)	2 (7%)
CHROMOPHOBE ADENOMA			2 (4%)	
#ADRENAL	(47)	(47)	(48)	(46)
PHEOCHROMOCYTOMA	7 (15%)	7 (15%)	7 (15%)	16 (35%)
PHEOCHROMOCYTOMA, MALIGNANT		1 (2%)		
#THYROID	(46)	(48)	(41)	(44)
FOLLICULAR-CELL ADENOMA				1 (2%)
C-CELL ADENOMA	2 (4%)			2 (5%)
C-CELL CARCINOMA	2 (4%)	1 (2%)	2 (5%)	2 (5%)
#PARATHYROID	(24)	(28)	(19)	(22)
ADENOMA, NOS	1 (4%)	1 (4%)		
#PANCREATIC ISLETS	(45)	(46)	(47)	(45)
ISLET-CELL ADENOMA	3 (7%)		4 (9%)	1 (2%)
ISLET-CELL CARCINOMA	1 (2%)			
REPRODUCTIVE SYSTEM				
*PREPUTIAL GLAND	(47)	(48)	(50)	(49)
CARCINOMA, NOS				1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-005	HIGH DOSE 01-0092
ADENOMA, NOS	1 (2%)		1 (2%)	2 (4%)
*SEMINAL VESICLE	(47)	(48)	(50)	(49)
MUCINOUS ADENOCARCINOMA, METASTA	1 (2%)			
#TESTIS	(47)	(47)	(49)	(48)
INTERSTITIAL-CELL TUMOR	44 (94%)	42 (89%)	46 (94%)	43 (90%)
NERVOUS SYSTEM				
#BRAIN	(45)	(48)	(49)	(46)
SARCOMA, NOS, METASTATIC				1 (2%)
GLIOMA, NOS		1 (2%)	1 (2%)	
OLIGODENDROGLIOMA			1 (2%)	
SPECIAL SENSE ORGANS				
*EAR CANAL	(47)	(48)	(50)	(49)
SQUAMOUS CELL PAPILLOMA				1 (2%)
SQUAMOUS CELL CARCINOMA	1 (2%)		1 (2%)	
*ZYMBAL'S GLAND	(47)	(48)	(50)	(49)
SQUAMOUS CELL CARCINOMA			1 (2%)	5 (10%)
MUSCULOSKELETAL SYSTEM				
*STERNUM	(47)	(48)	(50)	(49)
MUCINOUS ADENOCARCINOMA, METASTA	1 (2%)			
BODY CAVITIES				
*BODY CAVITIES	(47)	(48)	(50)	(49)
MESOTHELIOMA, NOS			3 (6%)	5 (10%)
MESOTHELIOMA, MALIGNANT	2 (4%)	2 (4%)		
ALL OTHER SYSTEMS				
SITE UNKNOWN				
OSTEOSARCOMA				1

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A1 (CONCLUDED)

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
ANIMAL DISPOSITION SUMMARY				
ANIMALS INITIALLY IN STUDY	50	49	50	50
NATURAL DEATH@	2	6	7	15
MORIBUND SACRIFICE	8	8	6	6
SCHEDULED SACRIFICE	5	5	5	5
ACCIDENTALLY KILLED				
TERMINAL SACRIFICE	33	30	32	24
ANIMAL MISSING	2			
@ INCLUDES AUTOLYZED ANIMALS				
TUMOR SUMMARY				
TOTAL ANIMALS WITH PRIMARY TUMORS*	47	44	48	47
TOTAL PRIMARY TUMORS	96	80	99	137
TOTAL ANIMALS WITH BENIGN TUMORS	47	43	46	46
TOTAL BENIGN TUMORS	76	62	72	77
TOTAL ANIMALS WITH MALIGNANT TUMORS	11	17	14	37
TOTAL MALIGNANT TUMORS	15	18	16	49
TOTAL ANIMALS WITH SECONDARY TUMORS#	1	1	2	7
TOTAL SECONDARY TUMORS	6	1	2	8
TOTAL ANIMALS WITH TUMORS UNCERTAIN- BENIGN OR MALIGNANT	5		10	10
TOTAL UNCERTAIN TUMORS	5		11	11
TOTAL ANIMALS WITH TUMORS UNCERTAIN- PRIMARY OR METASTATIC				
TOTAL UNCERTAIN TUMORS				

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
\# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE A2
SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE RATS TREATED WITH HYDRAZOBENZENE

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
ANIMALS INITIALLY IN STUDY	50	50	50	50
ANIMALS MISSING	2			
ANIMALS NECROPSIED	48	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	47	50	50	50
INTEGUMENTARY SYSTEM				
*SKIN	(48)	(50)	(50)	(50)
BASAL-CELL CARCINOMA		1 (2%)		
*SUBCUTANEOUS TISSUE	(48)	(50)	(50)	(50)
FIBROMA		1 (2%)		
FIBROSARCOMA		1 (2%)		
RESPIRATORY SYSTEM				
#LUNG	(47)	(50)	(50)	(50)
SQUAMOUS CELL CARCINOMA			1 (2%)	
SQUAMOUS CELL CARCINOMA, METASTA		1 (2%)		
ADENOCARCINOMA, NOS, METASTATIC				1 (2%)
ALVEOLAR/BRONCHIOLAR ADENOMA		1 (2%)	2 (4%)	1 (2%)
LEIOMYOSARCOMA, METASTATIC				1 (2%)
HEMATOPOIETIC SYSTEM				
*MULTIPLE ORGANS	(48)	(50)	(50)	(50)
MALIGNANT LYMPHOMA, NOS	2 (4%)			
UNDIFFERENTIATED LEUKEMIA		1 (2%)		
MYELOMONOCYTIC LEUKEMIA	3 (6%)	3 (6%)		
#SPLEEN	(47)	(48)	(50)	(50)
PHEOCHROMOCYTOMA, METASTATIC				1 (2%)
UNDIFFERENTIATED LEUKEMIA		1 (2%)		
#LYMPH NODE	(40)	(47)	(44)	(31)
C-CELL CARCINOMA, METASTATIC				1 (3%)
#THYMUS	(39)	(34)	(38)	(24)
THYMOMA	1 (3%)			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABEL A2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
CIRCULATORY SYSTEM				
NONE				
DIGESTIVE SYSTEM				
#LIVER	(47)	(50)	(50)	(50)
NEOPLASTIC NODULE				6 (12%)
PHEOCHROMOCYTOMA, METASTATIC				1 (2%)
#STOMACH	(46)	(48)	(50)	(44)
SQUAMOUS CELL PAPILLOMA			1 (2%)	
#ILEUM	(46)	(48)	(50)	(40)
LEIOMYOSARCOMA		1 (2%)		
URINARY SYSTEM				
NONE				
ENDOCRINE SYSTEM				
#PITUITARY	(46)	(40)	(47)	(37)
CARCINOMA,NOS	2 (4%)			
ADENOMA, NOS	18 (39%)	17 (43%)	11 (23%)	13 (35%)
CHROMOPHOBE ADENOMA			10 (21%)	
#ADRENAL	(47)	(49)	(49)	(49)
CORTICAL ADENOMA	3 (6%)	1 (2%)	1 (2%)	
CORTICAL CARCINOMA	1 (2%)			
PHEOCHROMOCYTOMA	1 (2%)	3 (6%)	3 (6%)	2 (4%)
PHEOCHROMOCYTOMA, MALIGNANT				1 (2%)
#ADRENAL MEDULLA	(47)	(49)	(49)	(49)
GANGLIONEUROMA		1 (2%)		
#THYROID	(46)	(45)	(50)	(46)
FOLLICULAR-CELL CARCINOMA		1 (2%)		
C-CELL ADENOMA		1 (2%)	2 (4%)	
C-CELL CARCINOMA	3 (7%)	1 (2%)	3 (6%)	3 (7%)
#PANCREATIC ISLETS	(46)	(48)	(49)	(46)
ISLET-CELL ADENOMA		2 (4%)		

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
REPRODUCTIVE SYSTEM				
*MAMMARY GLAND	(48)	(50)	(50)	(50)
ADENOMA, NOS				2 (4%)
ADENOCARCINOMA, NOS	1 (2%)		3 (6%)	6 (12%)
PAPILLARY CYSTADENOMA, NOS	1 (2%)			
LEIOMYOSARCOMA				1 (2%)
FIBROADENOMA	14 (29%)	19 (38%)	9 (18%)	9 (18%)
*CLITORAL GLAND	(48)	(50)	(50)	(50)
CARCINOMA,NOS				1 (2%)
SQUAMOUS CELL PAPILLOMA		1 (2%)		
ADENOMA, NOS		2 (4%)		6 (12%)
*VAGINA	(48)	(50)	(50)	(50)
SARCOMA, NOS	1 (2%)			
#UTERUS	(47)	(50)	(50)	(48)
CARCINOMA,NOS			1 (2%)	
ADENOMA, NOS				1 (2%)
ADENOCARCINOMA, NOS	2 (4%)	1 (2%)	2 (4%)	2 (4%)
LEIOMYOSARCOMA			1 (2%)	
ENDOMETRIAL STROMAL POLYP	15 (32%)	10 (20%)	10 (20%)	5 (10%)
ENDOMETRIAL STROMAL SARCOMA		1 (2%)	3 (6%)	1 (2%)
#UTERUS/ENDOMETRIUM	(47)	(50)	(50)	(48)
CARCINOMA,NOS			1 (2%)	1 (2%)
ADENOCARCINOMA, NOS			1 (2%)	
#OVARY	(46)	(49)	(50)	(45)
GRANULOSA-CELL TUMOR		1 (2%)	1 (2%)	1 (2%)
GRANULOSA-CELL CARCINOMA	1 (2%)			
NERVOUS SYSTEM				
#BRAIN	(47)	(50)	(50)	(49)
GLIOMA, NOS			2 (4%)	1 (2%)
ASTROCYTOMA	1 (2%)			
OLIGODENDROGLIOMA				1 (2%)
SPECIAL SENSE ORGANS				
*ZYMBAL'S GLAND	(48)	(50)	(50)	(50)
SQUAMOUS CELL CARCINOMA			1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
SEBACEOUS ADENOCARCINOMA				2 (4%)
MUSCULOSKELETAL SYSTEM				
NONE				
BODY CAVITIES				
NONE				
ALL OTHER SYSTEMS				
SITE UNKNOWN				
SQUAMOUS CELL CARCINOMA		1		
ANIMAL DISPOSITION SUMMARY				
ANIMALS INITIALLY IN STUDY	50	50	50	50
NATURAL DEATH@	3	5	2	10
MORIBUND SACRIFICE	6	3	6	14
SCHEDULED SACRIFICE	5	5	5	5
ACCIDENTALLY KILLED				
TERMINAL SACRIFICE	34	37	37	21
ANIMAL MISSING	2			

@ INCLUDES AUTOLYZED ANIMALS

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A2 (CONCLUDED)

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
TUMOR SUMMARY				
TOTAL ANIMALS WITH PRIMARY TUMORS*	42	38	39	36
TOTAL PRIMARY TUMORS	70	73	69	66
TOTAL ANIMALS WITH BENIGN TUMORS	38	35	34	30
TOTAL BENIGN TUMORS	53	59	49	39
TOTAL ANIMALS WITH MALIGNANT TUMORS	14	12	18	17
TOTAL MALIGNANT TUMORS	17	13	19	20
TOTAL ANIMALS WITH SECONDARY TUMORS#		1		4
TOTAL SECONDARY TUMORS		1		5
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT		1	1	7
TOTAL UNCERTAIN TUMORS		1	1	7
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC				
TOTAL UNCERTAIN TUMORS				

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
\# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

APPENDIX B

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MICE TREATED WITH HYDRAZOBENZENE

TABLE B1
. SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE MICE TREATED WITH HYDRAZOBENZENE

	LOW DOSE CONTROL (UNTR) 05-0070	HIGH DOSE CONTROL (UNTR) 05-0118	LOW DOSE 05-0050	HIGH DOSE 05-0093
ANIMALS INITIALLY IN STUDY	50	50	50	50
ANIMALS MISSING		1	1	2
ANIMALS NECROPSIED	50	49	49	48
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	50	49	47	46
INTEGUMENTARY SYSTEM				
*SUBCUT TISSUE	(50)	(49)	(49)	(48)
SARCOMA, NOS				1 (2%)
RESPIRATORY SYSTEM				
#LUNG	(50)	(49)	(47)	(46)
NEOPLASM, NOS, METASTATIC			1 (2%)	
HEPATOCELLULAR CARCINOMA, METAST	1 (2%)	1 (2%)	1 (2%)	
ALVEOLAR/BRONCHIOLAR ADENOMA		5 (10%)	1 (2%)	3 (7%)
ALVEOLAR/BRONCHIOLAR CARCINOMA	5 (10%)	5 (10%)	1 (2%)	
HEMATOPOIETIC SYSTEM				
*MULTIPLE ORGANS	(50)	(49)	(49)	(48)
MALIGNANT LYMPHOMA, NOS	2 (4%)			
MALIG.LYMPHOMA, HISTIOCYTIC TYPE	1 (2%)	3 (6%)	1 (2%)	
UNDIFFERENTIATED LEUKEMIA				1 (2%)
LYMPHOCYTIC LEUKEMIA				1 (2%)
#SPLEEN	(50)	(49)	(47)	(44)
HEMANGIOSARCOMA		1 (2%)		
MALIG.LYMPHOMA, HISTIOCYTIC TYPE		1 (2%)		
#LYMPH NODE	(45)	(42)	(44)	(38)
MALIG.LYMPHOMA, HISTIOCYTIC TYPE	2 (4%)	1 (2%)	1 (2%)	
#MESENTERIC L. NODE	(45)	(42)	(44)	(38)
MALIG.LYMPHOMA, HISTIOCYTIC TYPE			1 (2%)	
#PEYERS PATCH	(49)	(49)	(47)	(42)
MALIG.LYMPHOMA, HISTIOCYTIC TYPE			1 (2%)	1 (2%)
CIRCULATORY SYSTEM				
NONE				

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE B1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 05-0070	HIGH DOSE CONTROL (UNTR) 05-0118	LOW DOSE 05-0050	HIGH DOSE 05-0093
DIGESTIVE SYSTEM				
#LIVER	(50)	(48)	(47)	(46)
HEPATOCELLULAR ADENOMA		2 (4%)		1 (2%)
HEPATOCELLULAR CARCINOMA	12 (24%)	6 (13%)	11 (23%)	8 (17%)
HEMANGIOMA	1 (2%)			
HEMANGIOSARCOMA, UNC PRIM OR MET		1 (2%)		
#JEJUNUM	(49)	(49)	(47)	(42)
ADENOCARCINOMA, NOS				1 (2%)
URINARY SYSTEM				
#KIDNEY	(49)	(49)	(47)	(46)
TUBULAR-CELL ADENOCARCINOMA			1 (2%)	
ENDOCRINE SYSTEM				
#ADRENAL	(49)	(44)	(45)	(42)
CORTICAL ADENOMA			1 (2%)	
PHEOCHROMOCYTOMA		1 (2%)	1 (2%)	3 (7%)
#THYROID	(40)	(45)	(45)	(36)
ADENOCARCINOMA, NOS	1 (3%)			
FOLLICULAR-CELL ADENOMA				1 (3%)
#PANCREATIC ISLETS	(46)	(47)	(47)	(44)
ISLET-CELL ADENOMA	1 (2%)		1 (2%)	
REPRODUCTIVE SYSTEM				
*PREPUTIAL GLAND	(50)	(49)	(49)	(48)
ADENOMA, NOS			1 (2%)	
#TESTIS	(50)	(48)	(47)	(46)
INTERSTITIAL-CELL TUMOR				1 (2%)
NERVOUS SYSTEM				
NONE				

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 05-0070	HIGH DOSE CONTROL (UNTR) 05-0118	LOW DOSE 05-0050	HIGH DOSE 05-0093
SPECIAL SENSE ORGANS				
*HARDERIAN GLAND	(50)	(49)	(49)	(48)
ADENOMA, NOS	1 (2%)			
MUSCULOSKELETAL SYSTEM				
NONE				
BODY CAVITIES				
NONE				
ALL OTHER SYSTEMS				
*MULTIPLE ORGANS	(50)	(49)	(49)	(48)
NEUROFIBROSARCOMA	1 (2%)			
ANIMAL DISPOSITION SUMMARY				
ANIMALS INITIALLY IN STUDY	50	50	50	50
NATURAL DEATH@	2		3	7
MORIBUND SACRIFICE			2	3
SCHEDULED SACRIFICE	5	10		5
ACCIDENTALLY KILLED				
TERMINAL SACRIFICE	43	39	44	33
ANIMAL MISSING		1	1	2

@ INCLUDES AUTOLYZED ANIMALS

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B1 (CONCLUDED)

	LOW DOSE CONTROL (UNTR) 05-0070	HIGH DOSE CONTROL (UNTR) 05-0118	LOW DOSE 05-0050	HIGH DOSE 05-0093
TUMOR SUMMARY				
TOTAL ANIMALS WITH PRIMARY TUMORS*	23	22	18	17
TOTAL PRIMARY TUMORS	27	26	22	22
TOTAL ANIMALS WITH BENIGN TUMORS	3	8	5	7
TOTAL BENIGN TUMORS	3	8	5	9
TOTAL ANIMALS WITH MALIGNANT TUMORS	22	15	15	13
TOTAL MALIGNANT TUMORS	24	17	17	13
TOTAL ANIMALS WITH SECONDARY TUMORS#	1	1	1	
TOTAL SECONDARY TUMORS	1	1	2	
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT				
TOTAL UNCERTAIN TUMORS				
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC		1		
TOTAL UNCERTAIN TUMORS		1		

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
\# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE B2
SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE MICE TREATED WITH HYDRAZOBENZENE

	LOW DOSE CONTROL (UNTR) 06-0070	HIGH DOSE CONTROL (UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
ANIMALS INITIALLY IN STUDY	50	50	47	50
ANIMALS NECROPSIED	48	50	40	44
ANIMALS EXAMINED HISTOPATHOLOGICALLY **	47	50	39	43
INTEGUMENTARY SYSTEM				
*SKIN	(48)	(50)	(40)	(44)
SARCOMA, NOS				1 (2%)
*SUBCUT TISSUE	(48)	(50)	(40)	(44)
UNDIFFERENTIATED CARCINOMA			1 (3%)	
RESPIRATORY SYSTEM				
#LUNG	(46)	(50)	(38)	(40)
HEPATOCELLULAR CARCINOMA, METAST	1 (2%)			
ALVEOLAR/BRONCHIOLAR ADENOMA		2 (4%)	2 (5%)	2 (5%)
ALVEOLAR/BRONCHIOLAR CARCINOMA	2 (4%)	1 (2%)	1 (3%)	
OSTEOSARCOMA, METASTATIC	1 (2%)			
HEMATOPOIETIC SYSTEM				
*MULTIPLE ORGANS	(48)	(50)	(40)	(44)
MALIGNANT LYMPHOMA, NOS	2 (4%)	2 (4%)	2 (5%)	1 (2%)
MALIG.LYMPHOMA, HISTIOCYTIC TYPE			5 (13%)	2 (5%)
UNDIFFERENTIATED LEUKEMIA				1 (2%)
LYMPHOCYTIC LEUKEMIA	1 (2%)			
ERYTHROCYTIC LEUKEMIA	1 (2%)			
#SPLEEN	(47)	(49)	(39)	(41)
HEMANGIOSARCOMA	1 (2%)			
MALIGNANT LYMPHOMA, NOS	1 (2%)			
MALIG.LYMPHOMA, HISTIOCYTIC TYPE				1 (2%)
#MESENTERIC L. NODE	(36)	(44)	(34)	(33)
MALIG.LYMPHOMA, HISTIOCYTIC TYPE	1 (3%)		1 (3%)	
#LIVER	(47)	(50)	(39)	(43)
MALIG.LYMPHOMA, HISTIOCYTIC TYPE				1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE B2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 06-0070	HIGH DOSE CONTROL (UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
#PEYERS PATCH	(45)	(48)	(39)	(37)
MALIGNANT LYMPHOMA, NOS	1 (2%)			
MALIG.LYMPHOMA, HISTIOCYTIC TYPE			2 (5%)	
CIRCULATORY SYSTEM				
NONE				
DIGESTIVE SYSTEM				
#LIVER	(47)	(50)	(39)	(43)
NEOPLASM, NOS				1 (2%)
HEPATOCELLULAR ADENOMA				2 (5%)
HEPATOCELLULAR CARCINOMA	2 (4%)	1 (2%)	4 (10%)	20 (47%)
URINARY SYSTEM				
NONE				
ENDOCRINE SYSTEM				
#PITUITARY	(43)	(42)	(34)	(9)
ADENOMA, NOS	5 (12%)	1 (2%)	3 (9%)	
CHROMOPHOBE ADENOMA		2 (5%)		
#ADRENAL	(47)	(48)	(39)	(40)
CORTICAL ADENOMA	1 (2%)			
PHEOCHROMOCYTOMA			1 (3%)	
REPRODUCTIVE SYSTEM				
#UTERUS	(43)	(47)	(34)	(33)
LEIOMYOMA	1 (2%)			
ENDOMETRIAL STROMAL POLYP			1 (3%)	
#CERVIX UTERI	(43)	(47)	(34)	(33)
SARCOMA, NOS				1 (3%)
#OVARY/OVIDUCT	(43)	(47)	(34)	(33)
PAPILLARY ADENOMA	1 (2%)	1 (2%)		

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 06-0070	HIGH DOSE CONTROL (UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
*OVARY	(45)	(48)	(37)	(31)
GRANULOSA-CELL TUMOR				1 (3%)
NERVOUS SYSTEM				
NONE				
SPECIAL SENSE ORGANS				
*HARDERIAN GLAND	(48)	(50)	(40)	(44)
ADENOMA, NOS	1 (2%)			
PAPILLARY ADENOMA		1 (2%)		
MUSCULOSKELETAL SYSTEM				
NONE				
BODY CAVITIES				
NONE				
ALL OTHER SYSTEMS				
OMENTUM				
HEMANGIOSARCOMA	1			
ANIMAL DISPOSITION SUMMARY				
ANIMALS INITIALLY IN STUDY	50	50	47	50
NATURAL DEATH@	6	2	8	14
MORIBUND SACRIFICE	3		2	3
SCHEDULED SACRIFICE	5	10		5
ACCIDENTALLY KILLED				2
TERMINAL SACRIFICE	36	38	37	26
ANIMAL MISSING				

@ INCLUDES AUTOLYZED ANIMALS

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B2 (CONCLUDED)

	LOW DOSE CONTROL (UNTR) 06-0070	HIGH DOSE CONTROL (UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
TUMOR SUMMARY				
TOTAL ANIMALS WITH PRIMARY TUMORS*	18	10	19	29
TOTAL PRIMARY TUMORS	22	11	23	34
TOTAL ANIMALS WITH BENIGN TUMORS	8	7	7	4
TOTAL BENIGN TUMORS	9	7	7	4
TOTAL ANIMALS WITH MALIGNANT TUMORS	12	4	15	25
TOTAL MALIGNANT TUMORS	13	4	16	28
TOTAL ANIMALS WITH SECONDARY TUMORS#	2			
TOTAL SECONDARY TUMORS	2			
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT				2
TOTAL UNCERTAIN TUMORS				2
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC				
TOTAL UNCERTAIN TUMORS				

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS
\# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

APPENDIX C

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN RATS TREATED WITH HYDRAZOBENZENE

TABLE C1
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS
TREATED WITH HYDRAZOBENZENE

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
ANIMALS INITIALLY IN STUDY	50	49	50	50
ANIMALS MISSING	2			
ANIMALS NECROPSIED	47	48	50	49
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	47	48	49	49
INTEGUMENTARY SYSTEM				
*SKIN	(47)	(48)	(50)	(49)
EPIDERMAL INCLUSION CYST	1 (2%)		1 (2%)	1 (2%)
INFLAMMATION, NECROTIZING	1 (2%)			
INFLAMMATION, ACUTE FOCAL	1 (2%)			
HYPERKERATOSIS			1 (2%)	
ACANTHOSIS			1 (2%)	1 (2%)
*SUBCUT TISSUE	(47)	(48)	(50)	(49)
FIBROMATOSIS				1 (2%)
METAPLASIA, OSSEOUS		1 (2%)		
RESPIRATORY SYSTEM				
#TRACHEA	(46)	(48)	(48)	(46)
INFLAMMATION, NOS		2 (4%)	4 (8%)	1 (2%)
#LUNG/BRONCHUS	(47)	(48)	(49)	(48)
BRONCHIECTASIS		1 (2%)		1 (2%)
INFLAMMATION, NOS		7 (15%)	1 (2%)	8 (17%)
INFLAMMATION, FOCAL			1 (2%)	
INFLAMMATION, NECROTIZING				2 (4%)
ABSCESS, NOS	1 (2%)			
#LUNG/BRONCHIOLE	(47)	(48)	(49)	(48)
INFLAMMATION, NOS			1 (2%)	
#LUNG	(47)	(48)	(49)	(48)
CONGESTION, NOS			1 (2%)	
HEMORRHAGE			1 (2%)	
INFLAMMATION, NOS				1 (2%)
INFLAMMATION, FOCAL			1 (2%)	
INFLAMMATION, INTERSTITIAL		4 (8%)	12 (24%)	16 (33%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE C1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
INFLAMMATION, NECROTIZING		1 (2%)		2 (4%)
ABSCESS, NOS				1 (2%)
PNEUMONIA, CHRONIC MURINE		1 (2%)		3 (6%)
FIBROSIS, FOCAL				1 (2%)
HYPERPLASIA, EPITHELIAL		1 (2%)	3 (6%)	1 (2%)
HYPERPLASIA, ALVEOLAR EPITHELIUM			1 (2%)	4 (8%)
HEMATOPOIETIC SYSTEM				
#BONE MARROW	(46)	(47)	(49)	(45)
MYELOFIBROSIS				1 (2%)
MEGAKARYOCYTOSIS			1 (2%)	
#SPLEEN	(47)	(48)	(49)	(48)
HEMORRHAGE				1 (2%)
INFLAMMATION, NOS				2 (4%)
FIBROSIS		1 (2%)		1 (2%)
NECROSIS, FOCAL				2 (4%)
INFARCT HEMORRHAGIC	1 (2%)			
HEMOSIDEROSIS		1 (2%)	3 (6%)	2 (4%)
HYPERPLASIA, HEMATOPOIETIC		9 (19%)	10 (20%)	8 (17%)
HYPERPLASIA, ERYTHROID		10 (21%)	23 (47%)	8 (17%)
HYPERPLASIA, RETICULUM CELL				1 (2%)
#LYMPH NODE	(42)	(44)	(48)	(27)
HEMORRHAGE		1 (2%)		1 (4%)
INFLAMMATION, NOS			1 (2%)	
HYPERPLASIA, NOS			2 (4%)	1 (4%)
RETICULOCYTOSIS			1 (2%)	
LYMPHOCYTOSIS			2 (4%)	
PLASMACYTOSIS		1 (2%)		
HYPERPLASIA, PLASMA CELL			1 (2%)	
HYPERPLASIA, LYMPHOID		3 (7%)	5 (10%)	1 (4%)
#PANCREATIC L.NODE	(42)	(44)	(48)	(27)
INFLAMMATION, ACUTE/CHRONIC	1 (2%)			
#ILEOCOLIC LYMPH NODE	(42)	(44)	(48)	(27)
LYMPHADENOPATHY	1 (2%)			
CIRCULATORY SYSTEM				
#MYOCARDIUM	(47)	(48)	(49)	(47)
INFLAMMATION, NOS			5 (10%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
INFLAMMATION, INTERSTITIAL		23 (48%)	39 (80%)	14 (30%)
FIBROSIS		12 (25%)	21 (43%)	23 (49%)
DEGENERATION, NOS	10 (21%)			
*AORTA	(47)	(48)	(50)	(49)
MINERALIZATION			1 (2%)	
*PULMONARY ARTERY	(47)	(48)	(50)	(49)
MINERALIZATION	1 (2%)			
DIGESTIVE SYSTEM				
#LIVER	(47)	(48)	(49)	(49)
FIBROSIS SEPTAL LIVER		2 (4%)		
DEGENERATION, HYALINE	1 (2%)			
NECROSIS, FOCAL	1 (2%)	2 (4%)	4 (8%)	2 (4%)
NECROSIS, COAGULATIVE			3 (6%)	
METAMORPHOSIS FATTY	3 (6%)		1 (2%)	10 (20%)
CYTOPLASMIC VACUOLIZATION			4 (8%)	
FOCAL CELLULAR CHANGE	12 (26%)			
HYPERPLASIA, FOCAL		15 (31%)	20 (41%)	8 (16%)
HYPERPLASIA, DIFFUSE			1 (2%)	2 (4%)
ANGIECTASIS		1 (2%)		3 (6%)
HEMATOPOIESIS				4 (8%)
#LIVER/CENTRILOBULAR	(47)	(48)	(49)	(49)
NECROSIS, NOS		1 (2%)		
*BILE DUCT	(47)	(48)	(50)	(49)
INFLAMMATION, NOS		3 (6%)	1 (2%)	
HYPERPLASIA, NOS	4 (9%)	43 (90%)	25 (50%)	21 (43%)
#PANCREAS	(45)	(46)	(47)	(45)
INFLAMMATION, NOS		17 (37%)	15 (32%)	7 (16%)
INFLAMMATION, ACUTE/CHRONIC	1 (2%)			
#PANCREATIC DUCT	(45)	(46)	(47)	(45)
HYPERPLASIA, NOS				1 (2%)
#PANCREATIC ACINUS	(45)	(46)	(47)	(45)
ATROPHY, NOS	3 (7%)		1 (2%)	
ATROPHY, FOCAL	1 (2%)			
HYPERTROPHY, FOCAL				1 (2%)
HYPERPLASIA, NOS	1 (2%)			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
HYPERPLASIA, FOCAL		1 (2%)	2 (4%)	
#ESOPHAGUS	(46)	(45)	(48)	(39)
DYSPLASIA, NOS		1 (2%)		
#STOMACH	(47)	(48)	(49)	(47)
INFLAMMATION, NOS		1 (2%)	4 (8%)	1 (2%)
INFLAMMATION, FOCAL			1 (2%)	
HYPERPLASIA, BASAL CELL		1 (2%)		4 (9%)
HYPERKERATOSIS		2 (4%)	1 (2%)	10 (21%)
ACANTHOSIS		2 (4%)	4 (8%)	17 (36%)
PARAKERATOSIS				1 (2%)
#GASTRIC MUCOSA	(47)	(48)	(49)	(47)
HYPERPLASIA, FOCAL				4 (9%)
#PEYERS PATCH	(47)	(46)	(48)	(39)
HYPERPLASIA, NOS		12 (26%)	8 (17%)	7 (18%)
#ILEUM	(47)	(46)	(48)	(39)
INFLAMMATION, NOS		2 (4%)		
#COLON	(46)	(46)	(47)	(33)
NEMATODIASIS			1 (2%)	
PARASITISM		3 (7%)		3 (9%)
URINARY SYSTEM				
#KIDNEY	(47)	(48)	(49)	(48)
CYST, NOS	1 (2%)			
GLOMERULONEPHRITIS, NOS		47 (98%)	48 (98%)	44 (92%)
INFLAMMATION, CHRONIC	39 (83%)			
FIBROSIS, DIFFUSE		6 (13%)		8 (17%)
#URINARY BLADDER	(47)	(43)	(46)	(40)
HYPERPLASIA, EPITHELIAL	1 (2%)	1 (2%)	3 (7%)	1 (3%)
HYPERPLASIA, DIFFUSE			1 (2%)	
ENDOCRINE SYSTEM				
#PITUITARY	(47)	(38)	(46)	(29)
HYPERPLASIA, NOS		1 (3%)		1 (3%)
HYPERPLASIA, FOCAL	3 (6%)	2 (5%)	2 (4%)	2 (7%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
#ADRENAL	(47)	(47)	(48)	(46)
HYPERPLASIA, NOS				1 (2%)
HYPERPLASIA, FOCAL	1 (2%)			
#ADRENAL CORTEX	(47)	(47)	(48)	(46)
HYPERPLASIA, FOCAL			1 (2%)	
#ADRENAL MEDULLA	(47)	(47)	(48)	(46)
HYPERPLASIA, NODULAR	3 (6%)	1 (2%)	7 (15%)	2 (4%)
HYPERPLASIA, FOCAL		4 (9%)		2 (4%)
#THYROID	(46)	(48)	(41)	(44)
FOLLICULAR CYST, NOS				2 (5%)
HYPERPLASIA, C-CELL	4 (9%)	3 (6%)	1 (2%)	
#PARATHYROID	(24)	(28)	(19)	(22)
HYPERPLASIA, NOS	1 (4%)	1 (4%)		
HYPERPLASIA, FOCAL				2 (9%)
#PANCREATIC ISLETS	(45)	(46)	(47)	(45)
HYPERPLASIA, NOS		1 (2%)	3 (6%)	1 (2%)
REPRODUCTIVE SYSTEM				
*MAMMARY GLAND	(47)	(48)	(50)	(49)
GALACTOCELE	1 (2%)	2 (4%)	4 (8%)	
HYPERPLASIA, NOS	1 (2%)	4 (8%)	11 (22%)	4 (8%)
*PREPUTIAL GLAND	(47)	(48)	(50)	(49)
INFLAMMATION, ACUTE	1 (2%)			
HYPERPLASIA, NOS				1 (2%)
#PROSTATE	(46)	(44)	(48)	(43)
INFLAMMATION, NOS		17 (39%)	25 (52%)	12 (28%)
INFLAMMATION, ACUTE	7 (15%)			
INFLAMMATION, ACUTE FOCAL	3 (7%)			
INFLAMMATION, ACUTE/CHRONIC	2 (4%)			
HYPERPLASIA, FOCAL			2 (4%)	1 (2%)
METAPLASIA, SQUAMOUS			1 (2%)	
#TESTIS	(47)	(47)	(49)	(48)
MINERALIZATION		1 (2%)		
DEGENERATION, NOS	1 (2%)			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1 (CONCLUDED)

	LOW DOSE CONTROL (UNTR) 01-0055	HIGH DOSE CONTROL (UNTR) 01-0118	LOW DOSE 01-0050	HIGH DOSE 01-0092
ATROPHY, NOS		6 (13%)	4 (8%)	6 (13%)
HYPERPLASIA, INTERSTITIAL CELL		3 (6%)	7 (14%)	3 (6%)
#TESTIS/TUBULE	(47)	(47)	(49)	(48)
MINERALIZATION			1 (2%)	2 (4%)
DEGENERATION, NOS	1 (2%)		3 (6%)	
NERVOUS SYSTEM				
#BRAIN	(45)	(48)	(49)	(46)
HEMORRHAGE			1 (2%)	
NECROSIS, NOS			1 (2%)	
SPECIAL SENSE ORGANS				
*EAR CANAL	(47)	(48)	(50)	(49)
METAPLASIA, SQUAMOUS			1 (2%)	
MUSCULOSKELETAL SYSTEM				
NONE				
BODY CAVITIES				
NONE				
ALL OTHER SYSTEMS				
OMENTUM				
NECROSIS, FAT		2		1
SPECIAL MORPHOLOGY SUMMARY				
ANIMAL MISSING/NO NECROPSY	2			
AUTO/NECROPSY/NO HISTO			1	
AUTOLYSIS/NO NECROPSY	1	1		1

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE RATS
TREATED WITH HYDRAZOBENZENE

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
ANIMALS INITIALLY IN STUDY	50	50	50	50
ANIMALS MISSING	2			
ANIMALS NECROPSIED	48	50	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY **	47	50	50	50
INTEGUMENTARY SYSTEM				
*SKIN	(48)	(50)	(50)	(50)
INFLAMMATION, NOS		1 (2%)		
*SUBCUT TISSUE	(48)	(50)	(50)	(50)
MINERALIZATION		1 (2%)		
ABSCESS, NOS		1 (2%)		
NECROSIS, NOS				1 (2%)
RESPIRATORY SYSTEM				
#TRACHEA	(47)	(49)	(50)	(47)
INFLAMMATION, NOS			5 (10%)	
#LUNG/BRONCHUS	(47)	(50)	(50)	(50)
INFLAMMATION, NOS		3 (6%)	2 (4%)	2 (4%)
INFLAMMATION, FOCAL			1 (2%)	
#LUNG	(47)	(50)	(50)	(50)
INFLAMMATION, FOCAL			2 (4%)	
INFLAMMATION, INTERSTITIAL		6 (12%)	29 (58%)	7 (14%)
PNEUMONIA, CHRONIC MURINE				1 (2%)
HYPERPLASIA, EPITHELIAL		1 (2%)		
HYPERPLASIA, ALVEOLAR EPITHELIUM			5 (10%)	1 (2%)
HEMATOPOIETIC SYSTEM				
#BONE MARROW	(45)	(46)	(49)	(46)
OSTEOSCLEROSIS		1 (2%)		
MYELOSCLEROSIS				1 (2%)
#SPLEEN	(47)	(48)	(50)	(50)
INFLAMMATION, NOS			6 (12%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE C2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
HEMOSIDEROSIS		12 (25%)	12 (24%)	9 (18%)
ERYTHROPHAGOCYTOSIS			1 (2%)	
HYPERPLASIA, HEMATOPOIETIC		25 (52%)	38 (76%)	21 (42%)
HYPERPLASIA, ERYTHROID		19 (40%)	40 (80%)	20 (40%)
HEMATOPOIESIS	1 (2%)			
#SPLENIC CAPSULE	(47)	(48)	(50)	(50)
HEMORRHAGIC CYST		1 (2%)		
#LYMPH NODE	(40)	(47)	(44)	(31)
HEMORRHAGE				1 (3%)
INFLAMMATION, NOS			4 (9%)	1 (3%)
HYPERPLASIA, NOS			1 (2%)	1 (3%)
RETICULOCYTOSIS			2 (5%)	4 (13%)
LYMPHOCYTOSIS			3 (7%)	4 (13%)
PLASMACYTOSIS		1 (2%)		
HYPERPLASIA, RETICULUM CELL			1 (2%)	
HYPERPLASIA, LYMPHOID		4 (9%)	12 (27%)	2 (6%)
CIRCULATORY SYSTEM				
#MYOCARDIUM	(47)	(50)	(50)	(50)
INFLAMMATION, NOS		1 (2%)		
INFLAMMATION, INTERSTITIAL		23 (46%)	41 (82%)	2 (4%)
FIBROSIS		15 (30%)	11 (22%)	17 (34%)
DEGENERATION, NOS	7 (15%)			
#ENDOCARDIUM	(47)	(50)	(50)	(50)
INFLAMMATION, NOS		1 (2%)		
DIGESTIVE SYSTEM				
#SALIVARY GLAND	(46)	(50)	(50)	(48)
HYPERPLASIA, INTRADUCTAL			1 (2%)	
#LIVER	(47)	(50)	(50)	(50)
DEGENERATION, NOS	1 (2%)			
NECROSIS, FOCAL		2 (4%)	6 (12%)	2 (4%)
NECROSIS, COAGULATIVE			1 (2%)	2 (4%)
METAMORPHOSIS FATTY	2 (4%)	6 (12%)	1 (2%)	10 (20%)
CYTOPLASMIC VACUOLIZATION			1 (2%)	
FOCAL CELLULAR CHANGE	25 (53%)			
HYPERPLASIA, FOCAL		38 (76%)	31 (62%)	29 (58%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
HYPERPLASIA, DIFFUSE			1 (2%)	3 (6%)
HYPERPLASIA, ERYTHROID		1 (2%)		
HEMATOPOIESIS		2 (4%)	5 (10%)	3 (6%)
*BILE DUCT	(48)	(50)	(50)	(50)
INFLAMMATION, NOS		1 (2%)	1 (2%)	
HYPERPLASIA, NOS	2 (4%)	32 (64%)	27 (54%)	24 (48%)
HYPERPLASIA, FOCAL		1 (2%)		1 (2%)
#PANCREAS	(46)	(48)	(49)	(46)
INFLAMMATION, NOS		6 (13%)	14 (29%)	6 (13%)
#PANCREATIC DUCT	(46)	(48)	(49)	(46)
HYPERPLASIA, NOS			1 (2%)	
#PANCREATIC ACINUS	(46)	(48)	(49)	(46)
ATROPHY, NOS	8 (17%)		1 (2%)	
HYPERTROPHY, FOCAL				2 (4%)
HYPERPLASIA, FOCAL			1 (2%)	
#STOMACH	(46)	(48)	(50)	(44)
INFLAMMATION, NOS		1 (2%)		
INFLAMMATION, FOCAL			1 (2%)	
HYPERPLASIA, BASAL CELL				2 (5%)
HYPERKERATOSIS			3 (6%)	2 (5%)
ACANTHOSIS		2 (4%)	6 (12%)	5 (11%)
#PEYERS PATCH	(46)	(48)	(50)	(40)
HYPERPLASIA, NOS		15 (31%)	8 (16%)	8 (20%)
#COLON	(45)	(46)	(49)	(37)
PARASITISM		2 (4%)	5 (10%)	4 (11%)
URINARY SYSTEM				
#KIDNEY	(47)	(50)	(50)	(50)
HYDRONEPHROSIS				1 (2%)
GLOMERULONEPHRITIS, NOS		43 (86%)	46 (92%)	37 (74%)
INFLAMMATION, INTERSTITIAL			3 (6%)	
INFLAMMATION, CHRONIC	29 (62%)			
FIBROSIS, FOCAL			1 (2%)	
FIBROSIS, DIFFUSE		1 (2%)		
POSTMORTEM CHANGE	1 (2%)			
#URINARY BLADDER	(47)	(46)	(46)	(38)
HYPERPLASIA, EPITHELIAL			2 (4%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
ENDOCRINE SYSTEM				
#PITUITARY	(46)	(40)	(47)	(37)
PERIVASCULITIS		1 (3%)		
HYPERPLASIA, FOCAL	1 (2%)	3 (8%)	2 (4%)	
#ADRENAL	(47)	(49)	(49)	(49)
METAMORPHOSIS FATTY		1 (2%)		
LIPOIDOSIS				1 (2%)
#ADRENAL CORTEX	(47)	(49)	(49)	(49)
CYST, NOS	1 (2%)			
DEGENERATION, NOS	3 (6%)			
METAMORPHOSIS FATTY	1 (2%)			
HYPERPLASIA, NODULAR	2 (4%)			
HYPERPLASIA, FOCAL	3 (6%)			1 (2%)
#ADRENAL MEDULLA	(47)	(49)	(49)	(49)
THROMBOSIS, NOS	1 (2%)			
HYPERPLASIA, NODULAR		3 (6%)	3 (6%)	
HYPERPLASIA, FOCAL	1 (2%)	3 (6%)		3 (6%)
#THYROID	(46)	(45)	(50)	(46)
CYSTIC FOLLICLES		1 (2%)		
HYPERPLASIA, PAPILLARY				1 (2%)
HYPERPLASIA, C-CELL	4 (9%)	1 (2%)	2 (4%)	1 (2%)
REPRODUCTIVE SYSTEM				
*MAMMARY GLAND	(48)	(50)	(50)	(50)
DILATATION/DUCTS	3 (6%)			
GALACTOCELE	7 (15%)	16 (32%)	18 (36%)	9 (18%)
HYPERPLASIA, NOS	4 (8%)	8 (16%)	20 (40%)	8 (16%)
*MAMMARY DUCT	(48)	(50)	(50)	(50)
FIBROSIS	2 (4%)			
*CLITORIS	(48)	(50)	(50)	(50)
NECROSIS, NOS				1 (2%)
*CLITORAL GLAND	(48)	(50)	(50)	(50)
HYPERPLASIA, NOS				1 (2%)
*VAGINA	(48)	(50)	(50)	(50)
INFLAMMATION, NOS				1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNTR) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
#UTERUS	(47)	(50)	(50)	(48)
HYDROMETRA	2 (4%)			
ABSCESS, NOS			3 (6%)	
HYPERPLASIA, ADENOMATOUS		1 (2%)		2 (4%)
HYPERPLASIA, STROMAL			1 (2%)	
METAPLASIA, SQUAMOUS			1 (2%)	
#UTERUS/ENDOMETRIUM	(47)	(50)	(50)	(48)
INFLAMMATION, NOS		22 (44%)	28 (56%)	16 (33%)
INFLAMMATION, SUPPURATIVE			2 (4%)	
INFLAMMATION, ACUTE	9 (19%)			
ABSCESS, NOS			2 (4%)	
INFLAMMATION, CHRONIC	1 (2%)			
HYPERPLASIA, NOS		6 (12%)	9 (18%)	9 (19%)
HYPERPLASIA, FOCAL			1 (2%)	
HYPERPLASIA, CYSTIC	3 (6%)		4 (8%)	4 (8%)
HYPERPLASIA, ADENOMATOUS		1 (2%)		
HYPERPLASIA, STROMAL	1 (2%)			
#OVARY/OVIDUCT	(47)	(50)	(50)	(48)
INFLAMMATION, NOS		10 (20%)	7 (14%)	1 (2%)
INFLAMMATION, SUPPURATIVE		2 (4%)	2 (4%)	
INFLAMMATION, ACUTE	3 (6%)			
INFLAMMATION ACTIVE CHRONIC	1 (2%)			
INFLAMMATION, CHRONIC	1 (2%)			
#OVARY	(46)	(49)	(50)	(45)
CYST, NOS		8 (16%)	10 (20%)	
INFLAMMATION, NOS			1 (2%)	
INFLAMMATION, CHRONIC	1 (2%)			
DEGENERATION, CYSTIC				1 (2%)
#OVARY/FOLLICLE	(46)	(49)	(50)	(45)
HYPERPLASIA, NOS	1 (2%)			
NERVOUS SYSTEM				
NONE				
SPECIAL SENSE ORGANS				
*EYE	(48)	(50)	(50)	(50)
CATARACT		1 (2%)		

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2 (CONCLUDED)

	LOW DOSE CONTROL (UNTR) 02-0055	HIGH DOSE CONTROL (UNIP) 02-0118	LOW DOSE 02-0052	HIGH DOSE 02-0091
*EYE/RETINA	(48)	(50)	(50)	(50)
ATROPHY, NOS		1 (2%)		
*HARDERIAN GLAND	(48)	(50)	(50)	(50)
HYPERPLASIA, NOS		1 (2%)		
MUSCULOSKELETAL SYSTEM				
*BONE	(48)	(50)	(50)	(50)
OSTEOSCLEROSIS	1 (2%)			
BODY CAVITIES				
*PLEURA	(48)	(50)	(50)	(50)
INFLAMMATION, CHRONIC	1 (2%)			
*EPICARDIUM	(48)	(50)	(50)	(50)
INFLAMMATION, CHRONIC	1 (2%)			
ALL OTHER SYSTEMS				
OMENTUM				
NECROSIS, FAT		1		
SPECIAL MORPHOLOGY SUMMARY				
ANIMAL MISSING/NO NECROPSY	2			
AUTO/NECROPSY/HISTO PERF				2
AUTO/NECROPSY/NO HISTO	1			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

APPENDIX D

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MICE TREATED WITH HYDRAZOBENZENE

TABLE D1
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE MICE
TREATED WITH HYDRAZOBENZENE

	LOW DOSE CONTROL (UNTR) 05-0070	HIGH DOSE CONTROL (UNTR) 05-0118	LOW DOSE 05-0050	HIGH DOSE 05-0093
ANIMALS INITIALLY IN STUDY	50	50	50	50
ANIMALS MISSING		1	1	2
ANIMALS NECROPSIED	50	49	49	48
ANIMALS EXAMINED HISTOPATHOLOGICALLY **	50	49	47	46
INTEGUMENTARY SYSTEM				
*SKIN	(50)	(49)	(49)	(48)
INFLAMMATION, NOS		1 (2%)	1 (2%)	
ULCER, NOS			1 (2%)	
INFLAMMATION, FOCAL		3 (6%)		
INFLAMMATION, NECROTIZING		1 (2%)		
ABSCESS, NOS	2 (4%)			
ACANTHOSIS			1 (2%)	
*SUBCUT TISSUE	(50)	(49)	(49)	(48)
ABSCESS, NOS			1 (2%)	
NECROSIS, NOS			2 (4%)	
NECROSIS, FAT	1 (2%)			
METAPLASIA, OSSEOUS				1 (2%)
RESPIRATORY SYSTEM				
#LUNG/BRONCHUS	(50)	(49)	(47)	(46)
INFLAMMATION, FOCAL		1 (2%)		
#LUNG/BRONCHIOLE	(50)	(49)	(47)	(46)
INFLAMMATION, NOS	1 (2%)			
INFLAMMATION, FOCAL		1 (2%)		
PERIVASCULITIS	1 (2%)			
#LUNG	(50)	(49)	(47)	(46)
HEMORRHAGE	2 (4%)			
INFLAMMATION, NOS				1 (2%)
INFLAMMATION, INTERSTITIAL		10 (20%)	3 (6%)	5 (11%)
HYPERPLASIA, ALVEOLAR EPITHELIUM	2 (4%)			
HEMATOPOIETIC SYSTEM				
#SPLEEN	(50)	(49)	(47)	(44)
HYPERPLASIA, NOS		6 (12%)	14 (30%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE D1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 05-0070	HIGH DOSE CONTROL (UNTR) 05-0118	LOW DOSE 05-0050	HIGH DOSE 05-0093
RETICULOCYTOSIS		1 (2%)		
HYPERPLASIA, HEMATOPOIETIC		5 (10%)	6 (13%)	4 (9%)
HYPERPLASIA, ERYTHROID			3 (6%)	
HYPERPLASIA, LYMPHOID	1 (2%)	1 (2%)		3 (7%)
#SPLENIC FOLLICLES	(50)	(49)	(47)	(44)
HYPERPLASIA, NOS	2 (4%)			
#HEMOLYMPH NODES	(50)	(49)	(47)	(44)
INFLAMMATION, NOS			2 (4%)	
#LYMPH NODE	(45)	(42)	(44)	(38)
HEMORRHAGE			4 (9%)	1 (3%)
INFLAMMATION, NOS		10 (24%)	15 (34%)	5 (13%)
HYPERPLASIA, NOS		1 (2%)	1 (2%)	1 (3%)
RETICULOCYTOSIS		2 (5%)	5 (11%)	1 (3%)
LYMPHOCYTOSIS			5 (11%)	
HYPERPLASIA, HEMATOPOIETIC			1 (2%)	
HYPERPLASIA, LYMPHOID		3 (7%)	5 (11%)	3 (8%)
#MESENTERIC L. NODE	(45)	(42)	(44)	(38)
HYPERPLASIA, RETICULUM CELL	1 (2%)			
CIRCULATORY SYSTEM				
#HEART	(49)	(49)	(47)	(44)
MINERALIZATION		1 (2%)		
#HEART/VENTRICLE	(49)	(49)	(47)	(44)
MELANIN				2 (5%)
#AORTIC VALVE	(49)	(49)	(47)	(44)
INFLAMMATION, ACUTE/CHRONIC	1 (2%)			
*ARTERY	(50)	(49)	(49)	(48)
INFLAMMATION, NOS				2 (4%)
DIGESTIVE SYSTEM				
#SALIVARY GLAND	(49)	(48)	(46)	(45)
PERIVASCULITIS	1 (2%)			
#LIVER	(50)	(48)	(47)	(46)
NECROSIS, FOCAL	1 (2%)	9 (19%)	5 (11%)	10 (22%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 05-0070	HIGH DOSE CONTROL (UNTR) 05-0118	LOW DOSE 05-0050	HIGH DOSE 05-0093
NECROSIS, COAGULATIVE			1 (2%)	1 (2%)
METAMORPHOSIS FATTY	2 (4%)		1 (2%)	
HEPATOCYTOMEGALY	2 (4%)			
DEPLETION	1 (2%)			
HYPERPLASIA, NODULAR	2 (4%)			
HYPERPLASTIC NODULE		1 (2%)	3 (6%)	
HYPERPLASIA, FOCAL	1 (2%)		1 (2%)	2 (4%)
HYPERPLASIA, DIFFUSE	1 (2%)			
HEMATOPOIESIS			2 (4%)	
#LIVER/CENTRILOBULAR	(50)	(48)	(47)	(46)
NECROSIS, NOS	1 (2%)			
#LIVER/KUPFFER CELL	(50)	(48)	(47)	(46)
HYPERPLASIA, NOS	1 (2%)			
#PANCREAS	(46)	(47)	(47)	(44)
INFLAMMATION, NOS		1 (2%)	1 (2%)	3 (7%)
INFLAMMATION, FOCAL	1 (2%)			
#STOMACH	(49)	(48)	(47)	(44)
INFLAMMATION, NOS			5 (11%)	3 (7%)
INFLAMMATION, FOCAL		2 (4%)		1 (2%)
INFLAMMATION, NECROTIZING		1 (2%)		
HYPERPLASIA, FOCAL		1 (2%)		
HYPERKERATOSIS		1 (2%)	3 (6%)	1 (2%)
ACANTHOSIS		1 (2%)	4 (9%)	1 (2%)
#GASTRIC MUCOSA	(49)	(48)	(47)	(44)
INFLAMMATION, FOCAL	1 (2%)			
#PEYERS PATCH	(49)	(49)	(47)	(42)
HYPERPLASIA, NOS	1 (2%)	7 (14%)	3 (6%)	6 (14%)
#ILEUM	(49)	(49)	(47)	(42)
INFLAMMATION, NECROTIZING				1 (2%)
#COLON	(46)	(43)	(45)	(39)
GRANULOMA, NOS	1 (2%)			
PARASITISM		3 (7%)		1 (3%)
URINARY SYSTEM				
#KIDNEY	(49)	(49)	(47)	(46)
POLYCYSTIC KIDNEY			1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D1 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 05-0070	HIGH DOSE CONTROL (UNTR) 05-0118	LOW DOSE 05-0050	HIGH DOSE 05-0093
GLOMERULONEPHRITIS, NOS		2 (4%)	2 (4%)	1 (2%)
INFLAMMATION, INTERSTITIAL	3 (6%)	16 (33%)	12 (26%)	1 (2%)
INFLAMMATION, CHRONIC			1 (2%)	
#URINARY BLADDER	(47)	(48)	(47)	(45)
INFLAMMATION, NOS				2 (4%)
HYPERPLASIA, EPITHELIAL	1 (2%)	4 (8%)	1 (2%)	1 (2%)
ENDOCRINE SYSTEM				
#ADRENAL	(49)	(44)	(45)	(42)
HYPERPLASIA, NOS		3 (7%)		
#ADRENAL/CAPSULE	(49)	(44)	(45)	(42)
HYPERPLASIA, NOS		3 (7%)	2 (4%)	1 (2%)
#ADRENAL CORTEX	(49)	(44)	(45)	(42)
HYPERTROPHY, FOCAL				1 (2%)
#PANCREATIC ISLETS	(46)	(47)	(47)	(44)
HYPERPLASIA, NOS			1 (2%)	
REPRODUCTIVE SYSTEM				
*PREPUTIAL GLAND	(50)	(49)	(49)	(48)
ABSCESS, NOS		1 (2%)	2 (4%)	
NECROSIS, NOS			1 (2%)	
#PROSTATE	(49)	(49)	(41)	(41)
INFLAMMATION, NOS				1 (2%)
*SEMINAL VESICLE	(50)	(49)	(49)	(48)
MINERALIZATION			1 (2%)	
NECROSIS, NOS			1 (2%)	
#TESTIS/TUBULE	(50)	(48)	(47)	(46)
MINERALIZATION			1 (2%)	
*EPIDIDYMIS	(50)	(49)	(49)	(48)
INFLAMMATION, NOS		1 (2%)		
NERVOUS SYSTEM				
NONE				

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D1 (CONCLUDED)

	LOW DOSE CONTROL (UNTR) 05-0070	HIGH DOSE CONTROL (UNTR) 05-0118	LOW DOSE 05-0050	HIGH DOSE 05-0093
SPECIAL SENSE ORGANS				
NONE				
MUSCULOSKELETAL SYSTEM				
NONE				
BODY CAVITIES				
NONE				
ALL OTHER SYSTEMS				
ADIPOSE TISSUE				
INFLAMMATION, ACUTE		1		
OMENTUM				
NECROSIS, FAT		1		
SPECIAL MORPHOLOGY SUMMARY				
NO LESION REPORTED	12	5	3	9
ANIMAL MISSING/NO NECROPSY		1	1	2
AUTO/NECROPSY/NO HISTO			2	2

\# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE MICE
TREATED WITH HYDRAZOBENZENE

	LOW DOSE CONTROL (UNTR) 06-0070	HIGH DOSE CONTROL (UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
ANIMALS INITIALLY IN STUDY	50	50	47	50
ANIMALS NECROPSIED	48	50	40	44
ANIMALS EXAMINED HISTOPATHOLOGICALLY **	47	50	39	43
INTEGUMENTARY SYSTEM				
*SUBCUT TISSUE	(48)	(50)	(40)	(44)
ABSCESS, NOS		1 (2%)		
RESPIRATORY SYSTEM				
#LUNG/BRONCHUS	(46)	(50)	(38)	(40)
INFLAMMATION, FOCAL		1 (2%)		
#LUNG/BRONCHIOLE	(46)	(50)	(38)	(40)
INFLAMMATION, NOS	1 (2%)			
HYPERPLASIA, NOS		1 (2%)		
#LUNG	(46)	(50)	(38)	(40)
INFLAMMATION, INTERSTITIAL	1 (2%)	14 (28%)	5 (13%)	3 (8%)
HYPERPLASIA, ALVEOLAR EPITHELIUM				2 (5%)
HEMATOPOIETIC SYSTEM				
#BONE MARROW	(46)	(49)	(38)	(40)
MYELOFIBROSIS	1 (2%)			
#SPLEEN	(47)	(49)	(39)	(41)
HEMOSIDEROSIS			1 (3%)	
HYPERPLASIA, NOS		9 (18%)	13 (33%)	
HYPERPLASIA, HEMATOPOIETIC		6 (12%)	8 (21%)	3 (7%)
HYPERPLASIA, ERYTHROID			1 (3%)	
HYPERPLASIA, LYMPHOID	1 (2%)	2 (4%)		
#SPLENIC FOLLICLES	(47)	(49)	(39)	(41)
HYPERPLASIA, NOS	3 (6%)			
#HEMOLYMPH NODES	(47)	(49)	(39)	(41)
INFLAMMATION, NOS		2 (4%)	2 (5%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE D2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 06-0070	HIGH DOSE CONTROL (UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
HYPERPLASIA, NOS		1 (2%)		
#LYMPH NODE	(36)	(44)	(34)	(33)
HEMORRHAGE				1 (3%)
INFLAMMATION, NOS	1 (3%)	9 (20%)	5 (15%)	2 (6%)
HYPERPLASIA, NOS	1 (3%)	3 (7%)	1 (3%)	1 (3%)
RETICULOCYTOSIS		1 (2%)	4 (12%)	5 (15%)
LYMPHOCYTOSIS			4 (12%)	5 (15%)
HYPERPLASIA, HEMATOPOIETIC		1 (2%)	2 (6%)	1 (3%)
HYPERPLASIA, PLASMA CELL	1 (3%)			
HYPERPLASIA, LYMPHOID		4 (9%)		1 (3%)
#MEDIASTINAL L.NODE	(36)	(44)	(34)	(33)
PLASMACYTOSIS			1 (3%)	
#ABDOMINAL LYMPH NODE	(36)	(44)	(34)	(33)
PLASMACYTOSIS	1 (3%)			
CIRCULATORY SYSTEM				
#HEART	(44)	(50)	(39)	(42)
MINERALIZATION				1 (2%)
#HEART/VENTRICLE	(44)	(50)	(39)	(42)
MELANIN				2 (5%)
#MYOCARDIUM	(44)	(50)	(39)	(42)
INFLAMMATION, FOCAL		1 (2%)		
FIBROSIS, FOCAL	1 (2%)			
DIGESTIVE SYSTEM				
#SALIVARY GLAND	(45)	(48)	(38)	(38)
PERIVASCULITIS	3 (7%)			
PERIVASCULAR CUFFING	1 (2%)	3 (6%)		
#LIVER	(47)	(50)	(39)	(43)
MINERALIZATION				1 (2%)
INFLAMMATION, ACUTE FOCAL	1 (2%)			
INFLAMMATION, ACUTE/CHRONIC	1 (2%)			
NECROSIS, FOCAL	2 (4%)	7 (14%)	11 (28%)	3 (7%)
NECROSIS, COAGULATIVE				5 (12%)
HYPERPLASTIC NODULE			2 (5%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 06-0070	HIGH DOSE CONTROL (UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
HYPERPLASIA, FOCAL			1 (3%)	2 (5%)
HEMATOPOIESIS			3 (8%)	
*BILE DUCT	(48)	(50)	(40)	(44)
INFLAMMATION, ACUTE/CHRONIC	4 (8%)			
HYPERPLASIA, NOS				1 (2%)
#PANCREAS	(43)	(48)	(38)	(39)
INFLAMMATION, NOS	1 (2%)	2 (4%)	3 (8%)	
INFLAMMATION, INTERSTITIAL	1 (2%)			
PERIARTERITIS	1 (2%)			
#PANCREATIC ACINUS	(43)	(48)	(38)	(39)
ATROPHY, NOS	1 (2%)			
#STOMACH	(45)	(49)	(37)	(39)
INFLAMMATION, NOS		1 (2%)		
INFLAMMATION, FOCAL		1 (2%)	2 (5%)	
ULCER, FOCAL	1 (2%)			
HYPERKERATOSIS			4 (11%)	3 (8%)
ACANTHOSIS		2 (4%)	4 (11%)	2 (5%)
#PEYERS PATCH	(45)	(48)	(39)	(37)
HYPERPLASIA, NOS	1 (2%)	7 (15%)	1 (3%)	2 (5%)
URINARY SYSTEM				
#KIDNEY	(45)	(50)	(39)	(42)
MINERALIZATION				1 (2%)
GLOMERULONEPHRITIS, NOS	3 (7%)	4 (8%)	4 (10%)	1 (2%)
INFLAMMATION, NOS				1 (2%)
GLOMERULONEPHRITIS, FOCAL	2 (4%)	1 (2%)		
INFLAMMATION, INTERSTITIAL	1 (2%)	12 (24%)	9 (23%)	5 (12%)
GLOMERULONEPHRITIS, MEMBRANOUS	2 (4%)			
PYELONEPHRITIS, ACUTE/CHRONIC	1 (2%)			
GLOMERULONEPHRITIS, CHRONIC	1 (2%)			
#KIDNEY/GLOMERULUS	(45)	(50)	(39)	(42)
CYST, NOS				1 (2%)
#KIDNEY/TUBULE	(45)	(50)	(39)	(42)
MINERALIZATION		1 (2%)	1 (3%)	
DEGENERATION, CYSTIC				1 (2%)
#URINARY BLADDER	(45)	(48)	(37)	(41)
INFLAMMATION, CHRONIC FOCAL	1 (2%)			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 06-0070	HIGH DOSE CONTROL (UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
PERIARTERITIS	1 (2%)			
HYPERPLASIA, EPITHELIAL		1 (2%)	1 (3%)	1 (2%)
ENDOCRINE SYSTEM				
#ADRENAL/CAPSULE	(47)	(48)	(39)	(40)
NODULE			1 (3%)	1 (3%)
HYPERPLASIA, NOS		5 (10%)	2 (5%)	6 (15%)
#ADRENAL CORTEX	(47)	(48)	(39)	(40)
NODULE		1 (2%)	2 (5%)	
HYPERPLASIA, NOS		1 (2%)		
#THYROID	(41)	(44)	(34)	(33)
FOLLICULAR CYST, NOS				1 (3%)
INFLAMMATION, FOCAL		1 (2%)		
HYPERPLASIA, PAPILLARY		2 (5%)		
HYPERPLASIA, ADENOMATOUS		1 (2%)		
HYPERPLASIA, FOLLICULAR-CELL	1 (2%)			
#THYROID FOLLICLE	(41)	(44)	(34)	(33)
NECROSIS, FOCAL				1 (3%)
REPRODUCTIVE SYSTEM				
*MAMMARY GLAND	(48)	(50)	(40)	(44)
HYPERPLASIA, NOS		1 (2%)		
#UTERUS	(43)	(47)	(34)	(33)
HYDROMETRA	3 (7%)	13 (28%)		1 (3%)
CYST, NOS			1 (3%)	
ABSCESS, NOS	2 (5%)		3 (9%)	
METAPLASIA, SQUAMOUS			1 (3%)	
#UTERUS/ENDOMETRIUM	(43)	(47)	(34)	(33)
INFLAMMATION, NOS	2 (5%)	8 (17%)	2 (6%)	
INFLAMMATION, SUPPURATIVE	2 (5%)		2 (6%)	
INFLAMMATION, ACUTE	6 (14%)			
INFLAMMATION, ACUTE FOCAL	1 (2%)			
INFLAMMATION, ACUTE/CHRONIC	3 (7%)			
HYPERPLASIA, NOS	1 (2%)	8 (17%)	12 (35%)	
HYPERPLASIA, CYSTIC	20 (47%)	6 (13%)	8 (24%)	
METAPLASIA, SQUAMOUS	1 (2%)			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2 (CONTINUED)

	LOW DOSE CONTROL (UNTR) 06-0070	HIGH DOSE CONTROL (UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
#OVARY/OVIDUCT	(43)	(47)	(34)	(33)
INFLAMMATION, NOS		4 (9%)	1 (3%)	
INFLAMMATION, SUPPURATIVE	4 (9%)			
ABSCESS, NOS	1 (2%)	1 (2%)		
HYPERPLASIA, FOCAL			1 (3%)	
#OVARY	(45)	(48)	(37)	(31)
CYST, NOS		10 (21%)	5 (14%)	1 (3%)
INFLAMMATION, NOS		4 (8%)	6 (16%)	
INFLAMMATION, SUPPURATIVE	6 (13%)		4 (11%)	
ABSCESS, NOS			1 (3%)	
INFLAMMATION, CHRONIC	1 (2%)			
ABSCESS, CHRONIC	1 (2%)			
PERIARTERITIS	1 (2%)	1 (2%)		
DEGENERATION, CYSTIC		3 (6%)		
NERVOUS SYSTEM				
#BRAIN/MENINGES	(46)	(48)	(39)	(41)
INFLAMMATION, ACUTE/CHRONIC	1 (2%)			
INFLAMMATION, CHRONIC FOCAL	1 (2%)			
#BRAIN	(46)	(48)	(39)	(41)
NECROSIS, NOS				1 (2%)
SPECIAL SENSE ORGANS				
NONE				
MUSCULOSKELETAL SYSTEM				
NONE				
BODY CAVITIES				
NONE				
ALL OTHER SYSTEMS				
*MULTIPLE ORGANS	(48)	(50)	(40)	(44)
PERIVASCULITIS	1 (2%)			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2 (CONCLUDED)

	LOW DOSE CONTROL(UNTR) 06-0070	HIGH DOSE CONTROL(UNTR) 06-0118	LOW DOSE 06-0052	HIGH DOSE 06-0093
SPECIAL MORPHOLOGY SUMMARY				
NO LESION REPORTED		3		2
AUTO/NECROPSY/HISTO PERF		1		
AUTO/NECROPSY/NO HISTO	1		1	1
AUTOLYSIS/NO NECROPSY	2		7	6

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

Review of the Bioassay of Hydrazobenzene* for Carcinogenicity by the Data Evaluation/Risk Assessment Subgroup of the Clearinghouse on Environmental Carcinogens

The Clearinghouse on Environmental Carcinogens was established in May, 1976, in compliance with DHEW Committee Regulations and the Provisions of the Federal Advisory Committee Act. The purpose of the Clearinghouse is to advise the Director of the National Cancer Institute (NCI) on its bioassay program to identify and to evaluate chemical carcinogens in the environment to which humans may be exposed. The members of the Clearinghouse have been drawn from academia, industry, organized labor, public interest groups, State health officials, and quasi-public health and research organizations. Members have been selected on the basis of their experience in carcinogenesis or related fields and, collectively, provide expertise in chemistry, biochemistry, biostatistics, toxicology, pathology, and epidemiology. Representatives of various Governmental agencies participate as ad hoc members. The Data Evaluation/ Risk Assessment Subgroup of the Clearinghouse is charged with the responsibility of providing a peer review of reports prepared on NCI-sponsored bioassays of chemicals studied for carcinogenicity. It is in this context that the below critique is given on the bioassay of Hydrazobenzene for carcinogenicity.

The primary reviewer said that Hydrazobenzene was carcinogenic, under the conditions of test, in both sexes of treated rats and in treated female mice. He opined that the experimental design was adequate. In his critique he noted the following: the test chemical was obtained from a small laboratory and thus may not represent the chemical usually found in commerce; the stability of the compound in the diet mixture was not determined; the results of the subchronic study were not useful in establishing the dose levels for the chronic phase; and the high and low doses were too far apart and varied during the course of the chronic phase. Despite these shortcomings, the primary reviewer considered the study to be valid, given the unquestionable carcinogenic response induced in the treated animals. He said that Hydrazobenzene could pose a carcinogenic risk to humans.

The secondary reviewer concluded that, although Hydrazobenzene was quite toxic, it nevertheless was carcinogenic under the conditions of test. He noted that, in addition to the other tumors reported in the summary,

there was an increased incidence of adrenal tumors in several of the treated animal groups. He agreed that Hydrazobenzene may pose a carcinogenic risk to humans, even at low dose levels.

The primary reviewer reiterated his concern over the lack of stability data on Hydrazobenzene. He said that the compound could readily oxidize to azobenzene, a known hepatocarcinogen. A Subgroup member noted that, in any case, Hydrazobenzene probably is metabolized to azobenzene.

The primary reviewer moved that the report on the bioassay of Hydrazobenzene be accepted with the qualifications expressed in his critique. The motion was seconded and approved unanimously.

Members present were:

Michael Shimkin (Acting Chairman), University of California at San Diego
Joseph Highland, Environmental Defense Fund
George Roush, Jr., Monsanto Company
Louise Strong, University of Texas Health Sciences Center
John Weisburger, American Health Foundation

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* Subsequent to this review, changes may have been made in the bioassay report either as a result of the review or other reasons. Thus, certain comments and criticisms reflected in the review may no longer be appropriate.

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